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NEWS 22 APR 28 IMSRESEARCH reloaded with enhancements  
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NEWS 24 MAY 30 DGENE, PCTGEN, and USGENE enhanced with new homology  
sequence search option  
  
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FILE 'HOME' ENTERED AT 14:59:34 ON 04 JUN 2008

=> fil .bio  
COST IN U.S. DOLLARS SINCE FILE TOTAL

FULL ESTIMATED COST ENTRY SESSION  
0.21 0.21

FILE 'MEDLINE' ENTERED AT 14:59:49 ON 04 JUN 2008

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```
=> e endo y/au
E1      11      ENDO WATARU/AU
E2      2       ENDO WHALDENER/AU
E3      2561 --> ENDO Y/AU
E4      8       ENDO Y DR/AU
E5      2       ENDO Y I/AU
E6      1       ENDO Y M/AU
E7      424     ENDO YAETA/AU
E8      1       ENDO YAMAGAMI OSAMU/AU
E9      1       ENDO YASAHARU/AU
E10     1       ENDO YASHIRO/AU
E11     1       ENDO YASHUO/AU
E12     4       ENDO YASOICHI/AU
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=> s e3-e7
L1      2996 ("ENDO Y"/AU OR "ENDO Y DR"/AU OR "ENDO Y I"/AU OR "ENDO Y M"/AU OR
"ENDO YAETA"/AU)
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=> e sawasaki t/au
E1      1       SAWASAKI SUMIKO/AU
E2      2       SAWASAKI SUSUMU/AU
E3      91 --> SAWASAKI T/AU
E4      11      SAWASAKI TACHIO/AU
E5      3       SAWASAKI TAKA/AU
E6      1       SAWASAKI TAKAJI/AU
E7      24      SAWASAKI TAKASHI/AU
E8      11      SAWASAKI TAKESHI/AU
E9      3       SAWASAKI TAKURO/AU
E10     19      SAWASAKI TATSUO/AU
E11     201     SAWASAKI TATSUYA/AU
E12     1       SAWASAKI TATUYA/AU
```

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=> s e3
L2      91 "SAWASAKI T"/AU
```

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=> s e11-e12
L3      202 ("SAWASAKI TATSUYA"/AU OR "SAWASAKI TATUYA"/AU)
```

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=> s 11-13
L4      3093 (L1 OR L2 OR L3)
```

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=> s 14 AND ((protein(2a)synthes?) OR translation)(3a)((in(a)vitro) OR cell(a)free
OR cell-free)
L5      215 L4 AND ((PROTEIN(2A) SYNTHES?) OR TRANSLATION)(3A)((IN(A) VITRO) OR
```

CELL(A) FREE OR CELL-FREE)

=> dup rem l5

PROCESSING COMPLETED FOR L5

L6 121 DUP REM L5 (94 DUPLICATES REMOVED)

=> s l6 AND (mrna OR template OR wheat OR atp)

L7 103 L6 AND (MRNA OR TEMPLATE OR WHEAT OR ATP)

=> d ibib ed abs l7 1-103

L7 ANSWER 1 OF 103 MEDLINE on STN

ACCESSION NUMBER: 2008251993 MEDLINE [Full-text](#)

DOCUMENT NUMBER: PubMed ID: 18371187

TITLE: A set of ligation-independent in vitro translation vectors for eukaryotic protein production.

AUTHOR: Bardoczy Viola; Geczi Viktoria; Sawasaki Tatsuya; Endo Yasta; Meszaros Tamas

CORPORATE SOURCE: Budapest University of Technology and Economics, Department of Applied Biotechnology and Food Science, 1111 Budapest, Muegyetem rkp. 3., Hungary.. bardoczy@mail.bme.hu

SOURCE: BMC biotechnology, (2008) Vol. 8, pp. 32. Electronic Publication: 2008-03-27.

Journal code: 101088663. E-ISSN: 1472-6750.

PUB. COUNTRY: England; United Kingdom

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE) (RESEARCH SUPPORT, NON-U.S. GOV'T)

LANGUAGE: English

FILE SEGMENT: Priority Journals

ENTRY MONTH: 200805

ENTRY DATE: Entered STN: 17 Apr 2008

Last Updated on STN: 7 May 2008

Entered Medline: 6 May 2008

ED Entered STN: 17 Apr 2008

Last Updated on STN: 7 May 2008

Entered Medline: 6 May 2008

AB BACKGROUND: The last decade has brought the renaissance of protein studies and accelerated the development of high-throughput methods in all aspects of proteomics. Presently, most protein synthesis systems exploit the capacity of living cells to translate proteins, but their application is limited by several factors. A more flexible alternative protein production method is the cell-free in vitro protein translation. Currently available in vitro translation systems are suitable for high-throughput robotic protein production, fulfilling the requirements of proteomics studies. Wheat germ extract based in vitro translation system is likely the most promising method, since numerous eukaryotic proteins can be cost-efficiently synthesized in their native folded form. Although currently available vectors for wheat embryo in vitro translation systems ensure high productivity, they do not meet the requirements of state-of-the-art proteomics. Target genes have to be inserted using restriction endonucleases and the plasmids do not encode cleavable affinity purification tags. RESULTS: We designed four ligation independent cloning (LIC) vectors for wheat germ extract based in vitro protein translation. In these constructs, the RNA transcription is driven by T7 or SP6 phage polymerase and two TEV protease cleavable affinity tags can be added to aid protein purification. To evaluate our improved vectors, a plant mitogen activated protein kinase was cloned in all four constructs. Purification of this eukaryotic protein kinase demonstrated that all constructs functioned as intended: insertion of PCR fragment by LIC worked efficiently, affinity purification of translated proteins by GST-Sepharose or MagneHis particles resulted in high purity kinase, and the affinity tags could

efficiently be removed under different reaction conditions. Furthermore, high in vitro kinase activity testified of proper folding of the purified protein. CONCLUSION: Four newly designed in vitro translation vectors have been constructed which allow fast and parallel cloning and protein purification, thus representing useful molecular tools for high-throughput production of eukaryotic proteins.

L7 ANSWER 2 OF 103 MEDLINE on STN

ACCESSION NUMBER: 2008102820 MEDLINE [Full-text](#)

DOCUMENT NUMBER: PubMed ID: 18164779

TITLE: Production of yeast tRNA (m(7)G46) methyltransferase (Trm8-Trm82 complex) in a wheat germ cell-free translation system.

AUTHOR: Matsumoto Keisuke; Tomikawa Chie; Toyooka Takashi; Ochi Anna;

Takano Yoshitaka; Takayanagi Naoyuki; Abe Masato; Endo Yae; Hori Hiroyuki

CORPORATE SOURCE: Department of Applied Chemistry, Faculty of Engineering, Ehime University, Bunkyo 3, Matsuyama 790-8577, Japan.

SOURCE: Journal of biotechnology, (2008 Feb 29) Vol. 133, No. 4, pp.

453-60. Electronic Publication: 2007-11-24.

Journal code: 8411927. ISSN: 0168-1656.

PUB. COUNTRY: Netherlands

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
(RESEARCH SUPPORT, NON-U.S. GOV'T)

LANGUAGE: English

FILE SEGMENT: Priority Journals

ENTRY MONTH: 200805

ENTRY DATE: Entered STN: 12 Feb 2008

Last Updated on STN: 20 May 2008

Entered Medline: 19 May 2008

ED Entered STN: 12 Feb 2008

Last Updated on STN: 20 May 2008

Entered Medline: 19 May 2008

AB Cell-free translation systems are a powerful tool for the production of many kinds of proteins. However the production of proteins made up of hetero subunits is a major problem. In this study, we selected yeast tRNA (m(7)G46) methyltransferase (Trm8-Trm82 heterodimer) as a model protein. The enzyme catalyzes a methyl-transfer from S-adenosyl-L-methionine to the N(7) atom of guanine at position 46 in tRNA. When Trm8 or Trm82 mRNA were used for cell-free translation, Trm8 and Trm82 proteins could be synthesized. Upon mixing the synthesized Trm8 and Trm82 proteins, no active Trm8-Trm82 heterodimer was produced. Active Trm8-Trm82 heterodimer was only synthesized under conditions, in which both Trm8 and Trm82 mRNAs were co-translated. These results strongly suggest that the association of the Trm8 and Trm82 subunits is translationally controlled in living cells. Kinetic parameters of purified Trm8-Trm82 heterodimer were measured and these showed that the protein has comparable activity to other tRNA methyltransferases. The production of the m(7)G base at position 46 in tRNA was confirmed by two-dimensional thin layer chromatography and aniline cleavage of the methylated tRNA.

L7 ANSWER 3 OF 103 MEDLINE on STN

ACCESSION NUMBER: 2007735707 MEDLINE [Full-text](#)

DOCUMENT NUMBER: PubMed ID: 17981875

TITLE: A cell-free translation and proteoliposome reconstitution system for functional analysis of plant solute transporters.

AUTHOR: Nozawa Akira; Nanamiya Hideaki; Miyata Takuji; Linka Nicole;

Endo Yae; Weber Andreas P M; Tozawa Yuzuru

CORPORATE SOURCE: Cell-Free Science and Technology Research Center, and The Venture Business Laboratory, Ehime University, Matsuyama, Japan.

SOURCE: Plant & cell physiology, (2007 Dec) Vol. 48, No. 12, pp. 1815-

20. Electronic Publication: 2007-11-02.

Journal code: 9430925. ISSN: 0032-0781.

PUB. COUNTRY: Japan

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
(RESEARCH SUPPORT, NON-U.S. GOV'T)

LANGUAGE: English

FILE SEGMENT: Priority Journals

ENTRY MONTH: 200804

ENTRY DATE: Entered STN: 13 Dec 2007

Last Updated on STN: 2 Apr 2008

Entered Medline: 1 Apr 2008

ED Entered STN: 13 Dec 2007

Last Updated on STN: 2 Apr 2008

Entered Medline: 1 Apr 2008

AB We describe here a novel proteoliposome reconstitution system for functional analysis of plant membrane transporters that is based on a modified wheat germ cell-free translation system. We established optimized conditions for the reconstitution system with Arabidopsis thaliana phosphoenolpyruvate/phosphate translocator 1 (AtPPT1) as a model transporter. A high activity of AtPPT1 was achieved by synthesis of the protein in the presence of both a detergent such as Brij35 and liposomes. We also determined the substrate specificities of three putative rice PPT homologs with this system. The cell-free proteoliposome reconstitution system provides a valuable tool for functional analysis of transporter proteins.

L7 ANSWER 4 OF 103 MEDLINE on STN

ACCESSION NUMBER: 2007704683 MEDLINE [Full-text](#)

DOCUMENT NUMBER: PubMed ID: 18029735

TITLE: Hetero subunit interaction and RNA recognition of yeast tRNA (m7G46) methyltransferase synthesized in a wheat germ cell-free translation system.

AUTHOR: Muneyoshi Yuki; Matsumoto Keisuke; Tomikawa Chie; Toyooka

Takashi; Ochi Anna; Masaoka Takashi; Endo Yaeta; Hori Hiroyuki

CORPORATE SOURCE: Department of Materials and Biotechnology, Graduate School of Science and Engineering Ehime University, Matsuyama, 790-8577 Japan.

SOURCE: Nucleic acids symposium series (2004), (2007) No. 51, pp. 359-60.

Journal code: 101259965. E-ISSN: 1746-8272.

PUB. COUNTRY: England: United Kingdom

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
(RESEARCH SUPPORT, NON-U.S. GOV'T)

LANGUAGE: English

FILE SEGMENT: Priority Journals

ENTRY MONTH: 200804

ENTRY DATE: Entered STN: 29 Nov 2007

Last Updated on STN: 29 Apr 2008

Entered Medline: 28 Apr 2008

ED Entered STN: 29 Nov 2007

Last Updated on STN: 29 Apr 2008

Entered Medline: 28 Apr 2008

AB Yeast tRNA (m(7)G46) methyltransferase contains two protein subunits (Trm8 and Trm82). The enzyme catalyzes a methyl-transfer from S-adenosyl-L-methionine to the N(7) atom of guanine at position 46 in tRNA. We devised synthesis of active Trm8-Trm82 heterodimer in a wheat germ cell-free translation system. When Trm8 or Trm82 mRNA were used for a synthesis, Trm8 or Trm82 protein could be synthesized. Upon mixing the synthesized Trm8 and Trm82 proteins, no active Trm8-Trm82 heterodimer was produced. Active Trm8-Trm82 heterodimer was only synthesized under conditions, in which both Trm8 and Trm82 mRNAs were co-translated. To address the RNA recognition mechanism of the Trm8-Trm82 complex, we investigated methyl acceptance activities of eight truncated yeast

tRNA(Phe) transcripts. In this meeting, we demonstrate that yeast Trm8-Trm82 has stricter recognition requirements for the tRNA molecule as compared to the bacterial enzyme, TrmB.

L7 ANSWER 5 OF 103 MEDLINE on STN

ACCESSION NUMBER: 2007418268 MEDLINE [Full-text](#)  
DOCUMENT NUMBER: PubMed ID: 17634598  
TITLE: Methods for high-throughput materialization of genetic information based on wheat germ cell-free expression system.  
AUTHOR: Sawasaki Tatsuya; Morishita Ryo; Gouda Mudeppa D; Endo Yaeta  
CORPORATE SOURCE: Cell-Free Science and Technology Research Center, Ehime University, Matsuyama, Japan.  
SOURCE: Methods in molecular biology (Clifton, N.J.), (2007) Vol. 375, pp. 95-106. Ref: 10  
Journal code: 9214969. ISSN: 1064-3745.  
PUB. COUNTRY: United States  
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
General Review; (REVIEW)  
LANGUAGE: English  
FILE SEGMENT: Priority Journals  
ENTRY MONTH: 200708  
ENTRY DATE: Entered STN: 20 Jul 2007  
Last Updated on STN: 10 Aug 2007  
Entered Medline: 9 Aug 2007

ED Entered STN: 20 Jul 2007  
Last Updated on STN: 10 Aug 2007  
Entered Medline: 9 Aug 2007

AB Among the cell-free protein synthesis systems, the wheat germ-based translation system has significant advantages for the high-throughput production of eukaryotic multidomain proteins in folded state. Here, we describe protocols for this cell-free expression system.

L7 ANSWER 6 OF 103 MEDLINE on STN

ACCESSION NUMBER: 2007292338 MEDLINE [Full-text](#)  
DOCUMENT NUMBER: PubMed ID: 17367182  
TITLE: Construction of intramolecular luciferase complementation probe for detecting specific RNA.  
AUTHOR: Endoh Tamaki; Mie Masayasu; Funabashi Hisakage; Sawasaki Tatsuya; Endo Yaeta; Kobatake Eiry  
CORPORATE SOURCE: Department of Biological Information, Graduate School of Bioscience and Biotechnology, 4259, Nagatsuta, Yokohama, 226-8501, Japan.  
SOURCE: Bioconjugate chemistry, (2007 May-Jun) Vol. 18, No. 3, pp. 956-62. Electronic Publication: 2007-03-17.  
Journal code: 9010319. ISSN: 1043-1802.  
PUB. COUNTRY: United States  
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
(RESEARCH SUPPORT, NON-U.S. GOV'T)  
LANGUAGE: English  
FILE SEGMENT: Priority Journals  
ENTRY MONTH: 200708  
ENTRY DATE: Entered STN: 17 May 2007  
Last Updated on STN: 30 Aug 2007  
Entered Medline: 29 Aug 2007

ED Entered STN: 17 May 2007  
Last Updated on STN: 30 Aug 2007  
Entered Medline: 29 Aug 2007

AB Intermolecular enzyme complementation assay is a useful method for detecting protein-protein interactions. Specifically, bioluminescent signals produced

from reconstructed split luciferase fragments are powerful tools for in vivo analysis because the bioluminescent signals have been visualized both in cultured cells and living animals. However, they are limited for detection and evaluation of biological events relevant to intermolecular protein-protein interactions. In this study, we constructed an intramolecular luciferase complementation probe for detecting target biomolecules other than protein-protein interactions. It consists of peptide-inserted firefly luciferase (PI-FLuc) containing a short peptide between internally divided firefly luciferase. The inserted short peptide triggers FLuc complementation or discomplementation and subsequent reactivation or inactivation of FLuc activity through its induced fit conformational changes. We chose RNA binding arginine rich motif (ARM) peptides, Rev and/or Tat, for model peptide insertion, and expressed constructed PI-FLuc probe variants using a wheat germ cell-free protein synthesis system. They showed FLuc activity changes, reactivation, or inactivation after binding to their specific RNA targets. Furthermore, to expand the versatility of the PI-FLuc RNA detection system, we designed split-RNA probes built to reform the ARM peptide binding site in the presence of arbitrarily selected target-RNA. As a result, the target RNA was homogeneously detected by FLuc luminescent signals mediated by a cooperative function of the PI-FLuc and split-RNA probe sets.

L7 ANSWER 7 OF 103 MEDLINE on STN  
 ACCESSION NUMBER: 2007242104 MEDLINE [Full-text](#)  
 DOCUMENT NUMBER: PubMed ID: 17332011  
 TITLE: Novel protein fold discovered in the PabI family of restriction enzymes.  
 AUTHOR: Miyazono Ken-ichi; Watanabe Miki; Kosinski Jan; Ishikawa Ken; Kamo Masayuki; Sawasaki Tatsuya; Nagata Koji; Bujnicki Janusz M; Endo Yeeta; Tanokura Masaru; Kobayashi Ichizo  
 CORPORATE SOURCE: Department of Applied Biological Chemistry, Graduate School of Agricultural and Life Sciences, University of Tokyo, Tokyo, Japan.  
 SOURCE: Nucleic acids research, (2007) Vol. 35, No. 6, pp. 1908-18.  
 Electronic Publication: 2007-03-01.  
 Journal code: 0411011. E-ISSN: 1362-4962.  
 PUB. COUNTRY: England; United Kingdom  
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
 (RESEARCH SUPPORT, NON-U.S. GOV'T)  
 LANGUAGE: English  
 FILE SEGMENT: Priority Journals  
 OTHER SOURCE: PDB-2DVI  
 ENTRY MONTH: 200705  
 ENTRY DATE: Entered STN: 25 Apr 2007  
 Last Updated on STN: 8 May 2007  
 Entered Medline: 7 May 2007  
 ED Entered STN: 25 Apr 2007  
 Last Updated on STN: 8 May 2007  
 Entered Medline: 7 May 2007  
 AB Although structures of many DNA-binding proteins have been solved, they fall into a limited number of folds. Here, we describe an approach that led to the finding of a novel DNA-binding fold. Based on the behavior of Type II restriction-modification gene complexes as mobile elements, our earlier work identified a restriction enzyme, R.PabI, and its cognate modification enzyme in *Pyrococcus abyssi* through comparison of closely related genomes. While the modification methyltransferase was easily recognized, R.PabI was predicted to have a novel 3D structure. We expressed cytotoxic R.PabI in a wheat-germ-based cell-free translation system and determined its crystal structure. R.PabI turned out to adopt a novel protein fold. Homodimeric R.PabI has a curved anti-parallel beta-sheet that forms a 'half pipe'. Mutational and in silico DNA-binding analyses have assigned it as the double-strand DNA-binding

site. Unlike most restriction enzymes analyzed, R.PaBI is able to cleave DNA in the absence of Mg(2+). These results demonstrate the value of genome comparison and the wheat-germ-based system in finding a novel DNA-binding motif in mobile DNases and, in general, a novel protein fold in horizontally transferred genes.

L7 ANSWER 8 OF 103 MEDLINE on STN  
 ACCESSION NUMBER: 2007240109 MEDLINE Full-text  
 DOCUMENT NUMBER: PubMed ID: 17447497  
 TITLE: Wheat germ cell-free protein synthesis.  
 AUTHOR: Endo Yasta; Sawasaki Tatsuya  
 CORPORATE SOURCE: Cell-Free Science and Technology Research Center, Ehime University, Matsuyama, Japan.  
 SOURCE: Seikagaku. The Journal of Japanese Biochemical Society, (2007 Mar) Vol. 79, No. 3, pp. 229-38. Ref: 32  
 Journal code: 0413564. ISSN: 0037-1017.  
 PUB. COUNTRY: Japan  
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
 General Review; (REVIEW)  
 LANGUAGE: Japanese  
 FILE SEGMENT: Priority Journals  
 ENTRY MONTH: 200706  
 ENTRY DATE: Entered STN: 24 Apr 2007  
 Last Updated on STN: 13 Jun 2007  
 Entered Medline: 12 Jun 2007  
 ED Entered STN: 24 Apr 2007  
 Last Updated on STN: 13 Jun 2007  
 Entered Medline: 12 Jun 2007

L7 ANSWER 9 OF 103 MEDLINE on STN  
 ACCESSION NUMBER: 2007219087 MEDLINE Full-text  
 DOCUMENT NUMBER: PubMed ID: 17348022  
 TITLE: Detection of structural changes in a cofactor binding protein by using a wheat germ cell-free protein synthesis system coupled with unnatural amino acid probing.  
 AUTHOR: Abe Masato; Ohno Satoshi; Yokogawa Takashi; Nakanishi Takeshi; Arisaka Fumio; Hosoya Takamitsu; Hiramatsu Toshiyuki; Suzuki Masaaki; Ogasawara Tomio; Sawasaki Tatsuya; Nishikawa Kazuya; Kitamura Masaya; Hori Hiroyuki; Endo Yasta  
 CORPORATE SOURCE: Department of Applied Chemistry, Faculty of Engineering, Ehime University, Matsuyama 790-8577, Japan.  
 SOURCE: Proteins, (2007 May 15) Vol. 67, No. 3, pp. 643-52.  
 Journal code: 8700181. E-ISSN: 1097-0134.  
 PUB. COUNTRY: United States  
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
 (RESEARCH SUPPORT, NON-U.S. GOV'T)  
 LANGUAGE: English  
 FILE SEGMENT: Priority Journals  
 ENTRY MONTH: 200707  
 ENTRY DATE: Entered STN: 13 Apr 2007  
 Last Updated on STN: 10 Jul 2007  
 Entered Medline: 9 Jul 2007  
 ED Entered STN: 13 Apr 2007  
 Last Updated on STN: 10 Jul 2007  
 Entered Medline: 9 Jul 2007

AB A cell-free protein synthesis system is a powerful tool with which unnatural amino acids can be introduced into polypeptide chains. Here, the authors describe unnatural amino acid probing in a wheat germ cell-free translation system as a method for detecting the structural changes that occur in a



cofactor binding protein on a conversion of the protein from an apo-form to a holo-form. The authors selected the FMN-binding protein from *Desulfovibrio vulgaris* as a model protein. The apo-form of the protein was synthesized efficiently in the absence of FMN. The purified apo-form could be correctly converted to the holo-form. Thus, the system could synthesize the active apo-form. Gel filtration chromatography, analytical ultracentrifugation, and circular dichroism-spectra studies suggested that the FMN-binding site of the apo-form is open as compared with the holo-form. To confirm this idea, the unnatural amino acid probing was performed by incorporating 3-azido-L-tyrosine at the Tyr35 residue in the FMN-binding site. The authors optimized three steps in their system. The introduced 3-azido-L-tyrosine residue was subjected to specific chemical modification by a fluorescein-triarylphosphine derivative. The initial velocity of the apo-form reaction was 20 fold faster than that of the holo-form, demonstrating that the Tyr35 residue in the apo-form is open to solvent.

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L7 ANSWER 10 OF 103 MEDLINE on STN

ACCESSION NUMBER: 2007015216 MEDLINE Full-text

DOCUMENT NUMBER: PubMed ID: 17123829

TITLE: Sequence specificity and efficiency of protein N-terminal methionine elimination in wheat-embryo cell-free system.

AUTHOR: Kanno Takuya; Kitano Michiko; Kato Rika; Omori Akira; Endo Yaeta; Tozawa Yuzuru

CORPORATE SOURCE: Cell-Free Science and Technology Research Center, Ehime University, Matsuyama 790-8577, Japan.

SOURCE: Protein expression and purification, (2007 Mar) Vol. 52, No. 1, pp. 59-65. Electronic Publication: 2006-09-20.

Journal code: 9101496. ISSN: 1046-5928.

PUB. COUNTRY: United States

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English

FILE SEGMENT: Priority Journals

ENTRY MONTH: 200705

ENTRY DATE: Entered STN: 10 Jan 2007

Last Updated on STN: 10 May 2007

Entered Medline: 9 May 2007

ED Entered STN: 10 Jan 2007

Last Updated on STN: 10 May 2007

Entered Medline: 9 May 2007

AB Recent improvements in wheat-embryo cell-free translation resulted in a highly productive system for protein preparation. To clarify N-terminal processing of the cell-free system in a preparative-scale (> mg protein product per ml), 20 mutant variants of maltose-binding protein (MalE), each having a different penultimate residue in the sequence Met-Xaa-Ile-Glu-, and 20 glutathione S-transferase (GST) variants, having Met-Xaa-Pro-Ile-sequence, were designed and synthesized. The MalE and GST proteins were purified by amylose-resin and glutathione columns, respectively, followed by analysis of their N-terminal sequences. These investigations revealed that sequence specificity and efficiency of the N-terminal Met (N-Met) elimination in the cell-free system are similar to those reported from investigations in cellular systems or in the wheat-embryo cell-free protein expression system in analytical scale (approximately 10 microg protein product per ml). Cleavage of the N-Met is basically determined by the penultimate amino acid in the polypeptide sequence. In the case of MalE, the cleavage was efficient when the penultimate residue was Ala, Cys, Gly, Pro, Ser or Thr. But, in the case of GST with Pro as the antepenultimate residue, the efficiency was significantly reduced when the penultimate residue was Gly or Thr. We also confirmed that substitution of the antepenultimate residue in MalE to Pro drastically reduced

the efficiency of N-Met cleavage when the penultimate residue was Ala, Gly, Pro, Ser or Thr, indicating inhibitory effects of antepenultimate residue Pro on N-Met elimination. These results clarified sequence-specific functions of the endogenous N-terminal processing machinery in the scaled-up wheat-embryo cell-free translation system.

L7 ANSWER 11 OF 103 MEDLINE on STN  
 ACCESSION NUMBER: 2006472580 MEDLINE Full-text  
 DOCUMENT NUMBER: PubMed ID: 16828277  
 TITLE: Cell-free expression systems for eukaryotic protein production.  
 AUTHOR: Endo Yaeta; Sawasaki Tatsuya  
 CORPORATE SOURCE: Cell-Free Science and Technology Research Center, Ehime University, Matsuyama, Japan.. yendo@eng.ehime-u.ac.jp  
 SOURCE: Current opinion in biotechnology, (2006 Aug) Vol. 17, No. 4, pp. 373-80. Electronic Publication: 2006-07-07. Ref: 41  
 Journal code: 9100492. ISSN: 0958-1669.  
 PUB. COUNTRY: England: United Kingdom  
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
 (RESEARCH SUPPORT, NON-U.S. GOV'T)  
 General Review; (REVIEW)  
 LANGUAGE: English  
 FILE SEGMENT: Priority Journals  
 ENTRY MONTH: 200610  
 ENTRY DATE: Entered STN: 10 Aug 2006  
 Last Updated on STN: 6 Oct 2006  
 Entered Medline: 5 Oct 2006  
 ED Entered STN: 10 Aug 2006  
 Last Updated on STN: 6 Oct 2006  
 Entered Medline: 5 Oct 2006  
 AB Following the success of genome sequencing projects, attention has now turned to studies of the structure and function of proteins. Although cell-based expression systems for protein production have been widely used, they have certain limitations in terms of the quality and quantity of the proteins produced and for high-throughput production. Many of these limitations can be circumvented by the use of cell-free translation systems. Among such systems, the wheat germ based system is of special interest for its eukaryotic nature; it has the significant advantage of producing eukaryotic multidomain proteins in a folded state. Several advances in the use of cell-free expression systems have been made in the past few years and successful applications of these systems to produce proteins for functional and structural biology studies have been reported.

L7 ANSWER 12 OF 103 MEDLINE on STN  
 ACCESSION NUMBER: 2006375984 MEDLINE Full-text  
 DOCUMENT NUMBER: PubMed ID: 16708362  
 TITLE: Tolerance for random recombination of domains in prokaryotic and eukaryotic translation systems: Limited interdomain misfolding in a eukaryotic translation system.  
 AUTHOR: Hirano Nobutaka; Sawasaki Tatsuya; Tozawa Yuzuru; Endo Yaeta; Takai Kazuyuki  
 CORPORATE SOURCE: Venture Business Laboratory, Ehime University, Ehime, Japan.  
 SOURCE: Proteins, (2006 Aug 1) Vol. 64, No. 2, pp. 343-54.  
 Journal code: 8700181. E-ISSN: 1097-0134.  
 PUB. COUNTRY: United States  
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
 (RESEARCH SUPPORT, NON-U.S. GOV'T)  
 LANGUAGE: English  
 FILE SEGMENT: Priority Journals

ENTRY MONTH: 200608  
ENTRY DATE: Entered STN: 23 Jun 2006  
Last Updated on STN: 3 Aug 2006  
Entered Medline: 2 Aug 2006

ED Entered STN: 23 Jun 2006  
Last Updated on STN: 3 Aug 2006  
Entered Medline: 2 Aug 2006

AB It has been proposed that eukaryotic translation systems have a greater capacity for cotranslational folding of domains than prokaryotic translation systems, which reduces interdomain misfolding in multidomain proteins and, therefore, leads to tolerance for random recombination of domains. However, there has been a controversy as to whether prokaryotic and eukaryotic translation systems differ in the capacity for cotranslational domain folding. Here, to examine whether these systems differ in the tolerance for the random domain recombination, we systematically combined six proteins, out of which four are soluble and two are insoluble when produced in an *Escherichia coli* and a wheat germ cell-free protein synthesis systems, to construct a fusion protein library. Forty out of 60 two-domain proteins and 114 out of 120 three-domain proteins were more soluble when produced in the wheat system than in the *E. coli* system. Statistical analyses of the solubilities and the activities indicated that, in the wheat system but not in the *E. coli* system, the two soluble domains comprised mainly of beta-sheets tend to avoid interdomain misfolding and to fold properly even at the neighbor of the misfolded domains. These results demonstrate that a eukaryotic system permits the concomitance of a wider variety of domains within a single polypeptide chain than a prokaryotic system, which is probably due to the difference in the capacity for cotranslational folding. This difference is likely to be related to the postulated difference in the tolerance for random recombination of domains. Copyright 2006 Wiley-Liss, Inc.

L7 ANSWER 13 OF 103 MEDLINE on STN  
ACCESSION NUMBER: 2006307255 MEDLINE Full-text  
DOCUMENT NUMBER: PubMed ID: 16714002  
TITLE: In vitro selection of zinc finger DNA-binding proteins through  
ribosome display.  
AUTHOR: Ihara Hiroshi; Mie Masayasu; Funabashi Hisakage; Takahashi  
Fumio; Sawasaki Tatsuya; Endo Yaeta; Kobatake Eiry  
CORPORATE SOURCE: Department of Biological Information, Graduate School of  
Bioscience and Biotechnology, Tokyo Institute of Technology, 4259 Nagatsuta,  
Midoriku, Yokohama 226-8501, Japan.  
SOURCE: Biochemical and biophysical research communications, (2006 Jun  
7) Vol. 345, No. 3, pp. 1149-54. Electronic Publication: 2006-05-12.  
Journal code: 0372516. ISSN: 0006-291X.  
PUB. COUNTRY: United States  
DOCUMENT TYPE: (IN VITRO)  
Journal; Article; (JOURNAL ARTICLE)  
LANGUAGE: English  
FILE SEGMENT: Priority Journals  
ENTRY MONTH: 200608  
ENTRY DATE: Entered STN: 1 Jun 2006  
Last Updated on STN: 2 Aug 2006  
Entered Medline: 1 Aug 2006

ED Entered STN: 1 Jun 2006  
Last Updated on STN: 2 Aug 2006  
Entered Medline: 1 Aug 2006

AB DNA-binding proteins with sequence specificities have a variety of applications. To create novel functional DNA-binding proteins, in vivo selection methods have been developed. There are, however, crucial problems with such methods, e.g., limitation of library size and difficulty of

expression of toxic proteins for the host cells. In order to overcome these problems, we developed a novel way to select DNA-binding proteins using an in vitro ribosome display technique. The three zinc finger DNA-binding protein libraries, based on a Zif268 containing randomized sequence in each finger, were prepared and transcribed to mRNA in vitro. The ternary ribosomal complexes, formed by mRNA, ribosome, and translated DNA-binding protein during translation in a rabbit reticulocyte in vitro translation system, were selected with biotinylated target DNA fragments bound to streptavidin magnetic beads. The extracted mRNAs from the selected complexes were amplified using reverse transcription PCR and then sequenced. This is the first report of the selection of DNA-binding proteins involving an in vitro ribosome display technique.

L7 ANSWER 14 OF 103 MEDLINE on STN  
 ACCESSION NUMBER: 2005663866 MEDLINE Full-text  
 DOCUMENT NUMBER: PubMed ID: 16350953  
 TITLE: Advances in genome-wide protein expression using the wheat germ cell-free system.  
 AUTHOR: Endo Yasta; Sawasaki Tatsuya  
 CORPORATE SOURCE: Cell-Free Science and Technology Research Center, Ehime University, Matsuyama, Japan.  
 SOURCE: Methods in molecular biology (Clifton, N.J.), (2005) Vol. 310, pp. 145-67.  
 Journal code: 9214969. ISSN: 1064-3745.  
 PUB. COUNTRY: United States  
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
 LANGUAGE: English  
 FILE SEGMENT: Priority Journals  
 ENTRY MONTH: 200601  
 ENTRY DATE: Entered STN: 18 Dec 2005  
 Last Updated on STN: 24 Jan 2006  
 Entered Medline: 23 Jan 2006

ED Entered STN: 18 Dec 2005  
 Last Updated on STN: 24 Jan 2006  
 Entered Medline: 23 Jan 2006

AB In the current post-genomic era, cell-free translation platforms are gaining importance in structural as well as functional genomics. They are based on extracts prepared from Escherichia coli cells, wheat germ, or rabbit reticulocytes, and when programmed with any mRNA in the presence of energy sources and amino acids, can synthesize the respective protein in vitro. Among the cell-free systems, the wheat germ-based translation system is of special interest due to its eukaryotic nature and robustness. This chapter outlines the existing protein production platforms and their limitations, and describes the basic concept of the wheat germ-based cell-free system. It also demonstrates how the conventional wheat germ system can be improved by eliminating endogenous inhibitors, by using an expression vector specially designed for this system and polymerase chain reaction-directed protein synthesis directly from cDNAs in a bi-layer translation system. Finally, a robotic procedure for translation based on the wheat germ extract and bi-layer cell-free translation is described.

L7 ANSWER 15 OF 103 MEDLINE on STN  
 ACCESSION NUMBER: 2005634530 MEDLINE Full-text  
 DOCUMENT NUMBER: PubMed ID: 16213724  
 TITLE: Selection of 5'-untranslated sequences that enhance initiation of translation in a cell-free protein synthesis system from wheat embryos.  
 AUTHOR: Kamura Nami; Sawasaki Tatsuya; Kasahara Yuko; Takai Kazuyuki; Endo Yasta

CORPORATE SOURCE: Department of Applied Chemistry, Faculty of Engineering, Ehime University, 3, Bunkyo-cho, Matsuyama, Ehime 790-8577, Japan.  
SOURCE: Bioorganic & medicinal chemistry letters, (2005 Dec 15) Vol. 15, No. 24, pp. 5402-6. Electronic Publication: 2005-10-05.  
Journal code: 9107377. ISSN: 0960-894X.  
PUB. COUNTRY: England: United Kingdom  
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
(RESEARCH SUPPORT, NON-U.S. GOV'T)  
LANGUAGE: English  
FILE SEGMENT: Priority Journals  
ENTRY MONTH: 200606  
ENTRY DATE: Entered STN: 1 Dec 2005  
Last Updated on STN: 28 Jun 2006  
Entered Medline: 27 Jun 2006

ED Entered STN: 1 Dec 2005  
Last Updated on STN: 28 Jun 2006  
Entered Medline: 27 Jun 2006

AB Random libraries of mRNA 5'-leader sequences were screened to obtain some sequences that can stimulate the translation initiation in a cell-free translation system from wheat embryos as efficiently as the Omega sequence from tobacco mosaic virus. Several sequences that are as useful as the Omega sequence and are homologous to no known sequences survived the screening. We expect that these sequences add useful options to the cell-free protein synthesis system that is becoming a powerful tool in the post-genomic researches.

L7 ANSWER 16 OF 103 MEDLINE on STN

ACCESSION NUMBER: 2005536985 MEDLINE Full-text  
DOCUMENT NUMBER: PubMed ID: 16157309  
TITLE: Activity-based in vitro selection of T4 DNA ligase.  
AUTHOR: Takahashi Fumio; Funabashi Hisakage; Mie Masayasu; Endo Yaeta; Sawasaki Tatsuya; Aizawa Masuo; Kobatake Eiry  
CORPORATE SOURCE: Department of Biological Information, Graduate School of Bioscience and Biotechnology, Tokyo Institute of Technology, 4259 Nagatsuta, Midoriku, Yokohama 226-8501, Japan.  
SOURCE: Biochemical and biophysical research communications, (2005 Oct 28) Vol. 336, No. 3, pp. 987-93.  
Journal code: 0372516. ISSN: 0006-291X.

PUB. COUNTRY: United States  
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
LANGUAGE: English  
FILE SEGMENT: Priority Journals  
ENTRY MONTH: 200511  
ENTRY DATE: Entered STN: 12 Oct 2005  
Last Updated on STN: 3 Nov 2005  
Entered Medline: 1 Nov 2005

ED Entered STN: 12 Oct 2005  
Last Updated on STN: 3 Nov 2005  
Entered Medline: 1 Nov 2005

AB Recent in vitro methodologies for selection and directed evolution of proteins have concentrated not only on proteins with affinity such as single-chain antibody but also on enzymes. We developed a display technology for selection of T4 DNA ligase on ribosome because an in vitro selection method for DNA ligase had never been developed. The 3' end of mRNA encoding the gene of active or inactive T4 DNA ligase-spacer peptide fusion protein was hybridized to dsDNA fragments with cohesive ends, the substrate of T4 DNA ligase. After in vitro translation of the mRNA-dsDNA complex in a rabbit reticulocyte system, a mRNA-dsDNA-ribosome-ligase complex was produced. T4 DNA ligase enzyme displayed on a ribosome, through addition of a spacer peptide, is able

to react with dsDNA in the complex. The complex expressing active ligase was biotinylated by ligation with another biotinylated dsDNA probe and selected with streptavidin-coated magnetic beads. We effectively selected active T4 DNA ligase from a small amount of protein. The gene of the active T4 DNA ligase was enriched 40 times from a mixture of active and inactive genes using this selection strategy. This ribosomal display strategy may have high potential to be useful for selection of other enzymes associated with DNA.

L7 ANSWER 17 OF 103 MEDLINE on STN  
ACCESSION NUMBER: 2004567638 MEDLINE [Full-text](#)  
DOCUMENT NUMBER: PubMed ID: 15452433  
TITLE: A novel way of amino acid-specific assignment in (1)H-(15)N HSQC spectra with a wheat germ cell-free protein synthesis system.  
AUTHOR: Morita Eugene Hayato; Shimizu Masato; Ogasawara Tomio; Endo Yaeta; Tanaka Rikou; Kohno Toshiyuki  
CORPORATE SOURCE: Division of Gene Research, Department of Molecular Science, Integrated Center for Science, Ehime University, 3-5-7 Tarumi, Ehime 790-8566, Japan.. ehmorita@ipc.ehime-u.ac.jp  
SOURCE: Journal of biomolecular NMR, (2004 Sep) Vol. 30, No. 1, pp. 37-45.  
Journal code: 9110829. ISSN: 0925-2738.  
PUB. COUNTRY: Netherlands  
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
(RESEARCH SUPPORT, NON-U.S. GOV'T)  
LANGUAGE: English  
FILE SEGMENT: Priority Journals  
ENTRY MONTH: 200503  
ENTRY DATE: Entered STN: 16 Nov 2004  
Last Updated on STN: 12 Mar 2005  
Entered Medline: 11 Mar 2005  
ED Entered STN: 16 Nov 2004  
Last Updated on STN: 12 Mar 2005  
Entered Medline: 11 Mar 2005  
AB For high-throughput protein structural analyses, it is indispensable to develop a reliable protein overexpression system. Although many protein overexpression systems, such as ones utilizing E. coli cells, have been developed, a lot of proteins functioning in solution still were synthesized as insoluble forms. Recently, a novel wheat germ cell-free protein synthesis system was developed, and many of such proteins were synthesized as soluble forms. This means that the applicability of this protein synthesis method to determination of the functional structures of soluble proteins. In our previous work, we synthesized (15)N-labeled proteins with this wheat germ cell-free system, and confirmed this applicability on the basis of the strong similarity between the (1)H-(15)N HSQC spectra for native proteins and the corresponding ones for synthesized ones. In this study, we developed a convenient and reliable method for amino acid selective assignment in (1)H-(15)N HSQC spectra of proteins, using several inhibitors for transaminases and glutamine synthase in the process of protein synthesis. Amino acid selective assignment in (1)H-(15)N HSQC spectra is a powerful means to monitor the features of proteins, such as folding, intermolecular interactions and so on. This is also the first direct experimental evidence of the presence of active transaminases and glutamine synthase in wheat germ extracts.

L7 ANSWER 18 OF 103 MEDLINE on STN  
ACCESSION NUMBER: 2004466577 MEDLINE [Full-text](#)  
DOCUMENT NUMBER: PubMed ID: 15376967  
TITLE: In vitro protein synthesis system: cell-free protein synthesis system prepared from wheat germ.

AUTHOR: Sawasaki Tatsuya; Endo Yaetasasaki@eng.ehime-u.ac.jp  
SOURCE: Tanpakushitsu kakusan koso. Protein, nucleic acid, enzyme,  
(2004 Aug) Vol. 49, No. 11 Suppl, pp. 1514-9. Ref: 12  
Journal code: 0413762. ISSN: 0039-9450.

PUB. COUNTRY: Japan  
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
General Review; (REVIEW)

LANGUAGE: Japanese  
FILE SEGMENT: Priority Journals  
ENTRY MONTH: 200411  
ENTRY DATE: Entered STN: 21 Sep 2004  
Last Updated on STN: 3 Nov 2004  
Entered Medline: 2 Nov 2004

ED Entered STN: 21 Sep 2004  
Last Updated on STN: 3 Nov 2004  
Entered Medline: 2 Nov 2004

L7 ANSWER 19 OF 103 MEDLINE on STN  
ACCESSION NUMBER: 2004375184 MEDLINE [Full-text](#)  
DOCUMENT NUMBER: PubMed ID: 15276451  
TITLE: Genome-scale, biochemical annotation method based on the wheat  
germ cell-free protein synthesis system.

AUTHOR: Sawasaki Tatsuya; Hasegawa Yoshinori; Morishita Ryo; Seki  
Motoaki; Shinozaki Kazuo; Endo Yaeta  
CORPORATE SOURCE: Cell-Free Science and Technology Research Center, The Venture  
Business Laboratory, Ehime University, Matsuyama 790-8577, Japan.  
SOURCE: Phytochemistry, (2004 Jun) Vol. 65, No. 11, pp. 1549-55.  
Journal code: 0151434. ISSN: 0031-9422.

PUB. COUNTRY: United States  
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
(RESEARCH SUPPORT, NON-U.S. GOV'T)

LANGUAGE: English  
FILE SEGMENT: Priority Journals  
ENTRY MONTH: 200412  
ENTRY DATE: Entered STN: 28 Jul 2004  
Last Updated on STN: 19 Dec 2004  
Entered Medline: 7 Dec 2004

ED Entered STN: 28 Jul 2004  
Last Updated on STN: 19 Dec 2004  
Entered Medline: 7 Dec 2004

AB Since the complete genomic DNA sequencing of various species, attention has  
turned to the structural properties, and functional characteristics of  
proteins. Current cell-free protein expression systems from eukaryotes are  
capable of synthesizing proteins with high speed and accuracy; however, the  
yields are low due to their instability over time. This report reviews the  
high-throughput, genome-scale biochemical annotation method based on the cell-  
free system prepared from wheat embryos. We first briefly reviewed our highly  
efficient and robust wheat germ cell-free protein synthesis system, and then  
showed an application of the system for materialization and characterization  
of genetic information taking a cDNA library of protein kinase from  
*Arabidopsis thaliana* as an example. The procedure consists of: (1) fusion of  
the gene-of-interest to a purification-tag, amplified by the split-primer PCR  
method; (2) transcription and purification of mRNA; (3) cell-free protein  
synthesis in the bilayer system using 96-well titer plate; (4) affinity  
purification and activity measurement. We took 439 cDNAs encoding kinases  
among 1064 genes annotated so far, and they were translated in parallel into  
protein. Subsequent assay revealed 207 products having autophosphorylation  
activity. Furthermore, seven proteins out of 26 calcium-dependent protein  
kinase genes tested did phosphorylate a synthetic peptide substrate in the  
presence of calcium ion, demonstrating that the translation products, retained

their substrate specificity. The information on biochemical function of gene products accumulated should revolutionize our understanding of biology and fundamentally alter the practice of medicine and influence other industries as well.

L7 ANSWER 20 OF 103 MEDLINE on STN  
ACCESSION NUMBER: 2004151429 MEDLINE Full-text  
DOCUMENT NUMBER: PubMed ID: 15044017  
TITLE: Formation of circular polyribosomes in wheat germ cell-free protein synthesis system.  
AUTHOR: Madin Kairat; Sawasaki Tatsuya; Kamura Nami; Takai Kazuyuki; Ogasawara Tomio; Yazaki Kazumori; Takei Toshiaki; Miura Kin-Ichiro; Endo Yaeta  
CORPORATE SOURCE: Cell-free Science and Technology Research Center, and The Venture Business Laboratory, Ehime University, Matsuyama 790-8577, Japan.  
SOURCE: FEBS letters, (2004 Mar 26) Vol. 562, No. 1-3, pp. 155-9.  
Journal code: 0155157. ISSN: 0014-5793.  
PUB. COUNTRY: Netherlands  
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
LANGUAGE: English  
FILE SEGMENT: Priority Journals  
ENTRY MONTH: 200405  
ENTRY DATE: Entered STN: 27 Mar 2004  
Last Updated on STN: 12 May 2004  
Entered Medline: 11 May 2004

ED Entered STN: 27 Mar 2004  
Last Updated on STN: 12 May 2004  
Entered Medline: 11 May 2004

AB We report a morphological study of functioning ribosomes in a efficient and robust cell-free protein synthesis system prepared from wheat embryos. Sucrose density gradient analysis of translated mixtures programmed with luciferase mRNAs having different 5' and 3' untranslated regions showed formation of large polysomes. Electron microscopic examination of translation mixtures programmed with those of capped and polyadenylated mRNA revealed that ribosomes assemble into a circular-type polysome in vitro. Furthermore, a series of experiments using mRNAs lacking either cap, poly(A) tail or both also resulted in the formation of circular polysomes, which are indistinguishable from those with the original mRNA. The wheat germ cell-free system may provide a good experimental system for understanding functional ribosomes at the molecular level.

L7 ANSWER 21 OF 103 MEDLINE on STN  
ACCESSION NUMBER: 2003594215 MEDLINE Full-text  
DOCUMENT NUMBER: PubMed ID: 14675755  
TITLE: RALyase; a terminator of elongation function of depurinated ribosomes.  
AUTHOR: Ozawa Akihiko; Sawasaki Tatsuya; Takai Kazuyuki; Uchiyama Toshio; Hori Hiroyuki; Endo Yaeta  
CORPORATE SOURCE: Department of Applied Chemistry, Faculty of Engineering, Ehime University, Matsuyama, 790-8577, Japan.  
SOURCE: FEBS letters, (2003 Dec 18) Vol. 555, No. 3, pp. 455-8.  
Journal code: 0155157. ISSN: 0014-5793.  
PUB. COUNTRY: Netherlands  
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
LANGUAGE: English  
FILE SEGMENT: Priority Journals  
ENTRY MONTH: 200401  
ENTRY DATE: Entered STN: 17 Dec 2003  
Last Updated on STN: 24 Jan 2004



Entered Medline: 23 Jan 2004

ED Entered STN: 17 Dec 2003

Last Updated on STN: 24 Jan 2004

Entered Medline: 23 Jan 2004

AB Plant ribosomal RNA apurinic site specific lyase (RALYase) cleaves the phosphodiester bond at the depurinated site produced by ribosome-inactivating protein, while the biological role of this enzyme is not clear. As the depurinated ribosomes retain weak translation elongation activities, it was suggested that RALYase completes the ribosome inactivation. To confirm this point, we measured the effects of the phosphodiester cleavage using a fusion of wheat RALYase produced with a cell-free protein synthesis system from wheat germ. The results indicated that RALYase diminishes the residual elongation activities of the depurinated ribosomes.

L7 ANSWER 22 OF 103 MEDLINE on STN

ACCESSION NUMBER: 2003543983 MEDLINE Full-text

DOCUMENT NUMBER: PubMed ID: 14622267

TITLE: Efficient synthesis of a disulfide-containing protein through a batch cell-free system from wheat germ.

AUTHOR: Kawasaki Takayasu; Gouda Mudeppa D; Sawasaki Tatsuya; Takai Kazuyuki; Endo Yasta

CORPORATE SOURCE: Cell-free Science and Technology Research Center, Ehime University, Matsuyama, Japan.

SOURCE: European journal of biochemistry / FEBS, (2003 Dec) Vol. 270, No. 23, pp. 4780-6.

Journal code: 0107600. ISSN: 0014-2956.

PUB. COUNTRY: Germany: Germany, Federal Republic of

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
(RESEARCH SUPPORT, NON-U.S. GOV'T)

LANGUAGE: English

FILE SEGMENT: Priority Journals

ENTRY MONTH: 200401

ENTRY DATE: Entered STN: 19 Nov 2003

Last Updated on STN: 6 Jan 2004

Entered Medline: 5 Jan 2004

ED Entered STN: 19 Nov 2003

Last Updated on STN: 6 Jan 2004

Entered Medline: 5 Jan 2004

AB We have developed a highly productive cell-free protein synthesis system from wheat germ, which is expected to become an important tool for postgenomic research. However, this system has not been optimized for the synthesis of disulfide-containing proteins. Thus, we searched here for translation conditions under which a model protein, a single-chain antibody variable fragment (scFv), could be synthesized into its active form. Before the start of translation, the reducing agent dithiothreitol, which normally is added to the wheat germ extract but which inhibits disulfide formation during translation, was removed by gel filtration. When the scFv mRNA was incubated with this dithiothreitol-deficient extract, more than half of the synthesized polypeptide was recovered in the soluble fraction. By addition of protein disulfide isomerase in the translation solution, the solubility of the product was further improved, and nearly half of the soluble polypeptides strongly bound to the antigen immobilized on an agarose support. This strong binding component had a high affinity as shown by surface-plasmon resonance analysis. These results show that the wheat germ cell-free system can produce a functional scFv with a simple change of the reaction ingredients. We also discuss protein folding in this system and suggest that the disulfide bridges are formed cotranslationally. Finally, we show that biotinylated scFv could be synthesized in similar fashion and immobilized on a solid surface to which

streptavidin is bound. SPR measurements for detection of antigens were also possible with the use of this immobilized surface.

L7 ANSWER 23 OF 103 MEDLINE on STN  
ACCESSION NUMBER: 2003485228 MEDLINE Full-text  
DOCUMENT NUMBER: PubMed ID: 14563476  
TITLE: High-throughput, genome-scale protein production method based on the wheat germ cell-free expression system.  
AUTHOR: Endo Yasta; Sawasaki Tatsuya  
CORPORATE SOURCE: Department of Applied Chemistry, Faculty of Engineering, Ehime University, Matsuyama 790-8577, Japan.. yendo@en3.ehime-u.ac.jp  
SOURCE: Biotechnology advances, (2003 Nov) Vol. 21, No. 8, pp. 695-713.  
Ref: 39  
Journal code: 8403708. ISSN: 0734-9750.  
PUB. COUNTRY: England; United Kingdom  
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
General Review; (REVIEW)  
LANGUAGE: English  
FILE SEGMENT: Priority Journals  
ENTRY MONTH: 200407  
ENTRY DATE: Entered STN: 18 Oct 2003  
Last Updated on STN: 7 Jul 2004  
Entered Medline: 6 Jul 2004  
ED Entered STN: 18 Oct 2003  
Last Updated on STN: 7 Jul 2004  
Entered Medline: 6 Jul 2004

AB Cell-free protein synthesis systems can synthesize proteins with high speed and accuracy, but produce only a low yield because of their instability over time. Here we review our recent advances in a cell-free protein synthesis system prepared from wheat embryos. We first addressed and resolved the source of the instability of existing systems in light of endogenous ribosome-inactivating proteins. We found that conventional wheat germ extracts contained the RNA N-glycosidase tritin and other inhibitors such as thionin, ribonucleases, deoxyribonucleases, and proteases that originate from the endosperm and inhibit translation. Extensive washing of wheat embryos to eliminate endosperm contaminants has resulted in extracts with a high degree of stability and activity. To maximize the translation yield and throughput of the system, we then focused on developing the following issues: optimization of the ORF flanking regions, a new strategy to construct PCR-generated DNAs for screening, and design of an expression vector for large-scale protein production. The resulting system achieves high-throughput expression, with a PCR-directed system at least 50 genes that can be translated in parallel, yielding between 0.1 and 2.3 mg of protein by one person within 2 days. Under the dialysis mode of reaction, the system with the expression vector can maintain productive translation for 14 days. The cell-free system described here bypasses most of the biological processes and lends itself to robotic automation for high-throughput expression of genetic information, thus opening up many possibilities in the post-genome era.

L7 ANSWER 24 OF 103 MEDLINE on STN  
ACCESSION NUMBER: 2003369546 MEDLINE Full-text  
DOCUMENT NUMBER: PubMed ID: 12903243  
TITLE: Construction of an efficient expression vector for coupled transcription/translation in a wheat germ cell-free system.  
AUTHOR: Sawasaki Y; Hasegawa Y; Tsuchimochi M; Kasahara Y; Endo Y  
CORPORATE SOURCE: Department of Applied Chemistry, Faculty of Engineering, Ehime University, Matsuyama 790-8577, Japan.  
SOURCE: Nucleic acids symposium series, (2000) No. 44, pp. 9-10.

Journal code: 8007206. ISSN: 0261-3166.

PUB. COUNTRY: England; United Kingdom  
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
LANGUAGE: English  
FILE SEGMENT: Priority Journals  
ENTRY MONTH: 200310  
ENTRY DATE: Entered STN: 8 Aug 2003  
Last Updated on STN: 3 Oct 2003  
Entered Medline: 2 Oct 2003

ED Entered STN: 8 Aug 2003  
Last Updated on STN: 3 Oct 2003  
Entered Medline: 2 Oct 2003

AB Using the expression vector, pEU, which we have constructed, highly efficient in vitro protein synthesis can be achieved: The system works for 150 hours and without further template addition once the reaction has started, yielding 5 mg of enzymatically active protein in a 1 ml reaction.

L7 ANSWER 25 OF 103 MEDLINE on STN

ACCESSION NUMBER: 2003369492 MEDLINE Full-text  
DOCUMENT NUMBER: PubMed ID: 12903189  
TITLE: Identification of Aquifex aeolicus tRNA (m2(2G26) methyltransferase gene.  
AUTHOR: Takeda Hiroshi; Hori Hiroyuki; Endo Yaeta  
CORPORATE SOURCE: Department of Applied Chemistry, Faculty of Engineering, Ehime University, Matsuyama 790-8577, Japan.  
SOURCE: Nucleic acids research. Supplement (2001), (2002) No. 2, pp. 229-30.

Journal code: 101169367.

PUB. COUNTRY: England; United Kingdom  
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
(RESEARCH SUPPORT, NON-U.S. GOV'T)  
LANGUAGE: English  
FILE SEGMENT: Priority Journals  
ENTRY MONTH: 200309  
ENTRY DATE: Entered STN: 8 Aug 2003  
Last Updated on STN: 9 Sep 2003  
Entered Medline: 8 Sep 2003

ED Entered STN: 8 Aug 2003  
Last Updated on STN: 9 Sep 2003  
Entered Medline: 8 Sep 2003

AB The modifications of N2,N2-dimethylguanine (m2(2)G) are found in tRNAs and rRNAs from eukarya and archaea. In tRNAs, modification at position G26 is generated by tRNA (m2(2)G26) methyltransferase, which is encoded by the corresponding gene, trmI. This enzyme catalyzes the methyl-transfer from S-adenosyl-L-methionine to the semi-conserved residue, G26, via the intermediate modified base, m2G26. Recent genome sequencing project has been reported that the putative trmI is encoded in the genome of Aquifex aeolicus, a hyper-thermophilic eubacterium as only one exception among eubacteria. In order to confirm whether this bacterial trmI gene product is a real tRNA (m2(2)G26) methyltransferase or not, we expressed this protein by wheat germ in vitro cell-free translation system. Our biochemical analysis clearly showed that this gene product possessed tRNA (m2(2)G26) methyltransferase activity.

L7 ANSWER 26 OF 103 MEDLINE on STN

ACCESSION NUMBER: 2003356040 MEDLINE Full-text  
DOCUMENT NUMBER: PubMed ID: 12888530  
TITLE: Highly stable and efficient mRNA templates for mRNA-protein fusions and C-terminally labeled proteins.

AUTHOR: Miyamoto-Sato Etsuko; Takashima Hideaki; Fuse Shinichiro; Sue Kaori; Ishizaka Masamichi; Tateyama Seiji; Horisawa Kenichi; Sawasaki Tatsuya; Endo Tetsu; Yanagawa Hiroshi

CORPORATE SOURCE: Department of Biosciences and Informatics, Faculty of Science and Technology, Keio University, 3-14-1, Hiyoshi, Kohoku-ku, Yokohama 223-8522, Japan.

SOURCE: Nucleic acids research, (2003 Aug 1) Vol. 31, No. 15, pp. e78. Journal code: 0411011. E-ISSN: 1362-4962.

PUB. COUNTRY: England; United Kingdom

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
(RESEARCH SUPPORT, NON-U.S. GOV'T)

LANGUAGE: English

FILE SEGMENT: Priority Journals

ENTRY MONTH: 200308

ENTRY DATE: Entered STN: 31 Jul 2003

Last Updated on STN: 21 Aug 2003

Entered Medline: 20 Aug 2003

ED Entered STN: 31 Jul 2003

Last Updated on STN: 21 Aug 2003

Entered Medline: 20 Aug 2003

AB For high-throughput in vitro protein selection using genotype (mRNA)-phenotype (protein) fusion formation and C-terminal protein labeling as a post-selection analysis, it is important to improve the stability and efficiency of mRNA templates for both technologies. Here we describe an efficient single-strand ligation (90% of the input mRNAs) using a fluorescein-conjugated polyethylene glycol puromycin (Fluor-PEG Puro) spacer. This ligation provides a stable c-jun mRNA with a flexible Fluor-PEG Puro spacer for efficient fusion formation (70% of the input mRNA with the PEG spacer) in a cell-free wheat germ translation system. When using a 5' untranslated region including SP6 promoter and Omega29 enhancer (a part of tobacco mosaic virus Omega), an A(8) sequence (eight consecutive adenylate residues) at the 3' end is suitable for fusion formation, while an XA(8) sequence (XhoI and the A(8) sequence) is suitable for C-terminal protein labeling. Further, we report that Fluor-PEG N-t-butyloxycarbonylpuromycin (Puro(Boc)) spacer enhances the stability and efficiency of c-jun mRNA template for C-terminal protein labeling. These mRNA templates should be useful for puromycin-based technologies (fusion formation and C-terminal protein labeling) to facilitate high-throughput in vitro protein selection for not only evolutionary protein engineering, but also proteome exploration.

L7 ANSWER 27 OF 103 MEDLINE on STN

ACCESSION NUMBER: 2003258238 MEDLINE [Full-text](#)

DOCUMENT NUMBER: PubMed ID: 12761392

TITLE: A wheat germ cell-free system is a novel way to screen protein folding and function.

AUTHOR: Morita Eugene Hayato; Sawasaki Tatsuya; Tanaka Rikou; Endo Yae; Kohno Toshiyuki

CORPORATE SOURCE: Center for Gene Research, Ehime University, Ehime 790-8566, Japan.. ehmorita@ipc.ehime-u.ac.jp

SOURCE: Protein science : a publication of the Protein Society, (2003 Jun) Vol. 12, No. 6, pp. 1216-21.

Journal code: 9211750. ISSN: 0961-8368.

PUB. COUNTRY: United States

DOCUMENT TYPE: (COMPARATIVE STUDY)  
Journal; Article; (JOURNAL ARTICLE)  
(RESEARCH SUPPORT, NON-U.S. GOV'T)

LANGUAGE: English

FILE SEGMENT: Priority Journals

ENTRY MONTH: 200410

ENTRY DATE: Entered STN: 5 Jun 2003  
Last Updated on STN: 17 Dec 2003  
Entered Medline: 5 Oct 2004

ED Entered STN: 5 Jun 2003  
Last Updated on STN: 17 Dec 2003  
Entered Medline: 5 Oct 2004

AB For high-throughput protein structural analysis, it is indispensable to develop a reliable protein overexpression system. Although many protein overexpression systems, such as that involving *Escherichia coli* cells, have been developed, the number of overexpressed proteins showing the same biological activities as those of the native proteins is limited. A novel wheat germ cell-free protein synthesis system was developed recently, and most of the proteins functioning in solution were synthesized as soluble forms. This suggests the applicability of this protein synthesis method to determination of the solution structures of functional proteins. To examine this possibility, we have synthesized two (15)N-labeled proteins and obtained (1)H-(15)N HSQC spectra for them. The structural analysis of these proteins has already progressed with an *E. coli* overexpression system, and (1)H-(15)N HSQC spectra for biologically active proteins have already been obtained. Comparing the spectra, we have shown that proteins synthesized with a wheat germ cell-free system have the proper protein folding and enough biological activity. This is the first experimental evidence of the applicability of the wheat germ cell-free protein synthesis system to high-throughput protein structural analysis.

L7 ANSWER 28 OF 103 MEDLINE on STN  
ACCESSION NUMBER: 2003178039 MEDLINE [Full-text](#)  
DOCUMENT NUMBER: PubMed ID: 12696166  
TITLE: Recent advances in cell-free protein synthesis: application for postgenome sciences.  
AUTHOR: Endo Yaeta; Sawasaki Tatsuyayendo@eng.ehime-u.ac.jp  
SOURCE: Tanpakushitsu kakusan koso. Protein, nucleic acid, enzyme, (2003 Mar) Vol. 48, No. 4 Suppl, pp. 549-54. Ref: 12  
Journal code: 0413762. ISSN: 0039-9450.  
PUB. COUNTRY: Japan  
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
General Review; (REVIEW)  
LANGUAGE: Japanese  
FILE SEGMENT: Priority Journals  
ENTRY MONTH: 200306  
ENTRY DATE: Entered STN: 17 Apr 2003  
Last Updated on STN: 19 Jun 2003  
Entered Medline: 18 Jun 2003

ED Entered STN: 17 Apr 2003  
Last Updated on STN: 19 Jun 2003  
Entered Medline: 18 Jun 2003

L7 ANSWER 29 OF 103 MEDLINE on STN  
ACCESSION NUMBER: 2002669593 MEDLINE [Full-text](#)  
DOCUMENT NUMBER: PubMed ID: 12409616  
TITLE: A cell-free protein synthesis system for high-throughput proteomics.  
AUTHOR: Sawasaki Tatsuya; Ogasawara Tomio; Morishita Ryo; Endo Yaeta  
CORPORATE SOURCE: Department of Applied Chemistry, Faculty of Engineering, and Venture Business Laboratory, Ehime University, Matsuyama 790-8577, Japan.  
SOURCE: Proceedings of the National Academy of Sciences of the United States of America, (2002 Nov 12) Vol. 99, No. 23, pp. 14652-7. Electronic Publication: 2002-10-30.  
Journal code: 7505876. ISSN: 0027-8424.

PUB. COUNTRY: United States  
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
(RESEARCH SUPPORT, NON-U.S. GOV'T)  
LANGUAGE: English  
FILE SEGMENT: Priority Journals  
ENTRY MONTH: 200301  
ENTRY DATE: Entered STN: 14 Nov 2002  
Last Updated on STN: 16 Jan 2003  
Entered Medline: 15 Jan 2003

ED Entered STN: 14 Nov 2002  
Last Updated on STN: 16 Jan 2003  
Entered Medline: 15 Jan 2003

AB We report a cell-free system for the high-throughput synthesis and screening of gene products. The system, based on the eukaryotic translation apparatus of wheat seeds, has significant advantages over other commonly used cell-free expression systems. To maximize the yield and throughput of the system, we optimized the mRNA UTRs, designed an expression vector for large-scale protein production, and developed a new strategy to construct PCR-generated DNAs for high-throughput production of many proteins in parallel. The resulting system achieves high-yield expression and can maintain productive translation for 14 days. Additionally, in the integration of a PCR-directed system for template creation, at least 50 genes can be translated in parallel, yielding between 0.1 and 2.3 mg of protein by one person within 2 days. Assessment of correct protein folding by the products of this high-throughput protein-expression system were performed by enzymatic assays of kinases and by NMR spectroscopic analysis. The cell-free system, reported here, bypasses many of the time-consuming cloning steps of conventional expression systems and lends itself to a robotic automation for the high-throughput expression of proteins.

L7 ANSWER 30 OF 103 MEDLINE on STN

ACCESSION NUMBER: 2002397679 MEDLINE Full-text

DOCUMENT NUMBER: PubMed ID: 12097643

TITLE: An engineered Escherichia coli tyrosyl-tRNA synthetase for site-specific incorporation of an unnatural amino acid into proteins in eukaryotic translation and its application in a wheat germ cell-free system.

AUTHOR: Kiga Daisuke; Sakamoto Kensaku; Kodama Koichiro; Kigawa Takanori; Matsuda Takayoshi; Yabuki Takashi; Shirouzu Mikako; Harada Yoko; Nakayama Hiroshi; Takio Koji; Hasegawa Yoshinori; Endo Yaeta; Hirao Ichiro; Yokoyama Shigeyuki

CORPORATE SOURCE: RIKEN Genomic Sciences Center, 1-7-22 Suehiro-cho, Tsurumi, Yokohama 230-0045, Japan.

SOURCE: Proceedings of the National Academy of Sciences of the United States of America, (2002 Jul 23) Vol. 99, No. 15, pp. 9715-20. Electronic Publication: 2002-07-03.

Journal code: 7505876. ISSN: 0027-8424.

PUB. COUNTRY: United States  
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
LANGUAGE: English  
FILE SEGMENT: Priority Journals  
ENTRY MONTH: 200209  
ENTRY DATE: Entered STN: 31 Jul 2002  
Last Updated on STN: 5 Jan 2003  
Entered Medline: 4 Sep 2002

ED Entered STN: 31 Jul 2002  
Last Updated on STN: 5 Jan 2003  
Entered Medline: 4 Sep 2002

AB Tyrosyl-tRNA synthetase (TyrRS) from Escherichia coli was engineered to preferentially recognize 3-iodo-L-tyrosine rather than L-tyrosine for the site-specific incorporation of 3-iodo-L-tyrosine into proteins in eukaryotic

translation systems. The wild-type TyrRS does not recognize 3-iodo-L-tyrosine, because of the bulky iodine substitution. On the basis of the reported crystal structure of *Bacillus stearothermophilus* TyrRS, three residues, Y37, Q179, and Q195, in the L-tyrosine-binding site were chosen for mutagenesis. Thirty-four single amino acid replacements and 16 of their combinations were screened by in vitro biochemical assays. A combination of the Y37V and Q195C mutations changed the amino acid specificity in such a way that the variant TyrRS activates 3-iodo-L-tyrosine 10-fold more efficiently than L-tyrosine. This engineered enzyme, TyrRS(V37C195), was tested for use in the wheat germ cell-free translation system, which has recently been significantly improved, and is now as productive as conventional recombinant systems. During the translation in the wheat germ system, an *E. coli* suppressor tRNA(Tyr) was not aminoacylated by the wheat germ enzymes, but was aminoacylated by the *E. coli* TyrRS(V37C195) variant with 3-iodo-L-tyrosine. After the use of the 3-iodotyrosyl-tRNA in translation, the resultant uncharged tRNA could be aminoacylated again in the system. A mass spectrometric analysis of the produced protein revealed that more than 95% of the amino acids incorporated for an amber codon were idodotyrosine, whose concentration was only twice that of L-tyrosine in the translation. Therefore, the variant enzyme, 3-iodo-L-tyrosine, and the suppressor tRNA can serve as an additional set orthogonal to the 20 endogenous sets in eukaryotic in vitro translation systems.

L7 ANSWER 31 OF 103 MEDLINE on STN  
 ACCESSION NUMBER: 2002355633 MEDLINE [Full-text](#)  
 DOCUMENT NUMBER: PubMed ID: 12099014  
 TITLE: High-throughput expression of proteins from cDNAs catalogue from *Arabidopsis* in wheat germ cell-free protein synthesis system.  
 AUTHOR: Sawasaki Tatsuya; Seki Motoaki; Sinozaki Kazuo; Endo Yaetsawasasaki@en3.ehime-u.ac.jp  
 SOURCE: Tanpakushitsu kakusan koso. Protein, nucleic acid, enzyme, (2002 Jun) Vol. 47, No. 8 Suppl, pp. 1003-8. Ref: 16  
 Journal code: 0413762. ISSN: 0039-9450.  
 PUB. COUNTRY: Japan  
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
 General Review; (REVIEW)  
 LANGUAGE: Japanese  
 FILE SEGMENT: Priority Journals  
 ENTRY MONTH: 200208  
 ENTRY DATE: Entered STN: 9 Jul 2002  
 Last Updated on STN: 24 Aug 2002  
 Entered Medline: 23 Aug 2002  
 ED Entered STN: 9 Jul 2002  
 Last Updated on STN: 24 Aug 2002  
 Entered Medline: 23 Aug 2002

L7 ANSWER 32 OF 103 MEDLINE on STN  
 ACCESSION NUMBER: 2002290082 MEDLINE [Full-text](#)  
 DOCUMENT NUMBER: PubMed ID: 12030037  
 TITLE: Highly efficient cell-free protein synthesis system prepared from wheat embryos.  
 AUTHOR: Endo Yaetsa; Sawasaki Tatsuya  
 CORPORATE SOURCE: Department of Applied Chemistry, Faculty of Engineering, and Venture Business Laboratory, Ehime University, 3 Bunkyo-cho, Matsuyama 790-8577.  
 SOURCE: Seikagaku. The Journal of Japanese Biochemical Society, (2002 Apr) Vol. 74, No. 4, pp. 326-30. Ref: 8  
 Journal code: 0413564. ISSN: 0037-1017.  
 PUB. COUNTRY: Japan  
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
 General Review; (REVIEW)

LANGUAGE: Japanese  
FILE SEGMENT: Priority Journals  
ENTRY MONTH: 200207  
ENTRY DATE: Entered STN: 28 May 2002  
Last Updated on STN: 31 Jul 2002  
Entered Medline: 30 Jul 2002  
ED Entered STN: 28 May 2002  
Last Updated on STN: 31 Jul 2002  
Entered Medline: 30 Jul 2002

L7 ANSWER 33 OF 103 MEDLINE on STN  
ACCESSION NUMBER: 2002171983 MEDLINE Full-text  
DOCUMENT NUMBER: PubMed ID: 11904191  
TITLE: Ribosome display for selection of active dihydrofolate  
reductase mutants using immobilized methotrexate on agarose beads.  
AUTHOR: Takahashi Fumio; Ebihara Takashi; Mie Masayasu; Yanagida  
Yasuko; Endo Yaeta; Kobatake Eiry; Aizawa Masuo  
CORPORATE SOURCE: Department of Biological Information, Graduate School of  
Bioscience and Biotechnology, Tokyo Institute of Technology, 4259 Nagatsuta,  
Midoriku, 226-8501, Yokohama, Japan.  
SOURCE: FEBS letters, (2002 Mar 6) Vol. 514, No. 1, pp. 106-10.  
Journal code: 0155157. ISSN: 0014-5793.  
PUB. COUNTRY: Netherlands  
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
LANGUAGE: English  
FILE SEGMENT: Priority Journals  
ENTRY MONTH: 200204  
ENTRY DATE: Entered STN: 21 Mar 2002  
Last Updated on STN: 30 Jul 2002  
Entered Medline: 10 Apr 2002

ED Entered STN: 21 Mar 2002  
Last Updated on STN: 30 Jul 2002  
Entered Medline: 10 Apr 2002

AB Ribosome display was applied to the selection of an enzyme. As a model, we  
selected and amplified the dihydrofolate reductase (DHFR) gene by ribosome  
display utilizing a wheat germ cell-free protein synthesis system based on  
binding affinity to its substrate analog, methotrexate, immobilized on agarose  
beads. After three rounds of selection, the DHFR gene could be effectively  
selected and preferentially amplified from a small proportion in a mixture  
also containing competitive genes. Active enzymes were expressed and  
amplified and by sequence analysis, four mutants of DHFR were identified.  
These mutants showed as much activity as the wild-type enzyme.

L7 ANSWER 34 OF 103 MEDLINE on STN  
ACCESSION NUMBER: 2002171982 MEDLINE Full-text  
DOCUMENT NUMBER: PubMed ID: 11904190  
TITLE: A bilayer cell-free protein synthesis system for high-  
throughput screening of gene products.  
AUTHOR: Sawasaki Tatsuya; Hasegawa Yoshinori; Tsuchimochi Masateru;  
Kamura Nami; Ogasawara Tomio; Kuroita Toshihiro; Endo Yaeta  
CORPORATE SOURCE: Department of Applied Chemistry, Faculty of Engineering, The  
Venture Business Laboratory, Ehime University, 790-8577, Matsuyama, Japan.  
SOURCE: FEBS letters, (2002 Mar 6) Vol. 514, No. 1, pp. 102-5.  
Journal code: 0155157. ISSN: 0014-5793.  
PUB. COUNTRY: Netherlands  
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
LANGUAGE: English  
FILE SEGMENT: Priority Journals  
ENTRY MONTH: 200204



ENTRY DATE: Entered STN: 21 Mar 2002  
 Last Updated on STN: 12 Apr 2002  
 Entered Medline: 10 Apr 2002

ED Entered STN: 21 Mar 2002  
 Last Updated on STN: 12 Apr 2002  
 Entered Medline: 10 Apr 2002

AB A high-throughput cell-free protein synthesis method has been described. The methodology is based on a bilayer diffusion system that enables the continuous supply of substrates, together with the continuous removal of small byproducts, through a phase between the translation mixture and substrate mixture. With the use of a multititer plate the system was functional for a prolonged time, and as a consequence yielded more than 10 times that of the similar batch-mode reaction. Combining this method with a wheat germ cell-free translation system developed by us, the system could produce a large amount of protein sufficient for carrying out functional analyses. This novel bilayer-based cell-free protein synthesis system with its simplicity, minimum time and low cost may be useful practical methodology in the post-genome era.

L7 ANSWER 35 OF 103 MEDLINE on STN

ACCESSION NUMBER: 2000105511 MEDLINE Full-text  
 DOCUMENT NUMBER: PubMed ID: 10639118

TITLE: A highly efficient and robust cell-free protein synthesis system prepared from wheat embryos: plants apparently contain a suicide system directed at ribosomes.

AUTHOR: Madin K; Sawasaki T; Ogasawara T; Endo Y

CORPORATE SOURCE: Department of Applied Chemistry, Faculty of Engineering, Ehime University, Matsuyama 790-8577, Japan.

SOURCE: Proceedings of the National Academy of Sciences of the United States of America, (2000 Jan 18) Vol. 97, No. 2, pp. 559-64.  
 Journal code: 7505876. ISSN: 0027-8424.

PUB. COUNTRY: United States

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
 (RESEARCH SUPPORT, NON-U.S. GOV'T)

LANGUAGE: English

FILE SEGMENT: Priority Journals

ENTRY MONTH: 200003

ENTRY DATE: Entered STN: 14 Mar 2000  
 Last Updated on STN: 14 Mar 2000  
 Entered Medline: 2 Mar 2000

ED Entered STN: 14 Mar 2000  
 Last Updated on STN: 14 Mar 2000  
 Entered Medline: 2 Mar 2000

AB Current cell-free protein synthesis systems can synthesize proteins with high speed and accuracy, but produce only a low yield because of their instability over time. Here we describe the preparation of a highly efficient but also robust cell-free system from wheat embryos. We first investigated the source of the instability of existing systems in light of endogenous ribosome-inactivating proteins and found that ribosome inactivation by tritin occurs already during extract preparation and continues during incubation for protein synthesis. Therefore, we prepared our system from extensively washed embryos that are devoid of contamination by endosperm, the source of tritin and possibly other inhibitors. In a batch system, we observed continuous translation for 4 h, and sucrose density gradient analysis showed formation of large polysomes, indicating high protein synthesis activity. When the reaction was performed in a dialysis bag, enabling the continuous supply of substrates together with the continuous removal of small byproducts, translation proceeded for >60 h, yielding 1-4 mg of enzymatically active proteins, and 0.6 mg of a 126-kDa tobacco mosaic virus protein, per milliliter of reaction volume. Our results demonstrate that plants contain endogenous

inhibitors of translation and that after their elimination the translational apparatus is very stable. This contrasts with the common belief that cell-free translation systems are inherently unstable, even fragile. Our method is useful for the preparation of large amounts of active protein as well as for the study of protein synthesis itself.

L7 ANSWER 36 OF 103 MEDLINE on STN  
ACCESSION NUMBER: 1999329090 MEDLINE Full-text  
DOCUMENT NUMBER: PubMed ID: 10400702  
TITLE: Ribonuclease activity of rat liver perchloric acid-soluble protein, a potent inhibitor of protein synthesis.  
AUTHOR: Morishita R; Kawagoshi A; Sawasaki T; Madin K; Ogasawara T; Oka T; Endo Y  
CORPORATE SOURCE: Department of Applied Chemistry, Faculty of Engineering, Ehime University, Matsuyama 790-8577, Japan.  
SOURCE: The Journal of biological chemistry, (1999 Jul 16) Vol. 274, No. 29, pp. 20688-92.  
Journal code: 2985121R. ISSN: 0021-9258.  
PUB. COUNTRY: United States  
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
(RESEARCH SUPPORT, NON-U.S. GOV'T)  
LANGUAGE: English  
FILE SEGMENT: Priority Journals  
ENTRY MONTH: 199908  
ENTRY DATE: Entered STN: 27 Aug 1999  
Last Updated on STN: 27 Aug 1999  
Entered Medline: 19 Aug 1999

ED Entered STN: 27 Aug 1999  
Last Updated on STN: 27 Aug 1999  
Entered Medline: 19 Aug 1999  
AB Rat liver perchloric acid-soluble protein (L-PSP) is a potent inhibitor of cell-free protein synthesis; however, its mechanism of action is not known. Here we show that the protein is a unique ribonuclease and that this activity is responsible for the inhibition of translation. The addition of perchloric acid-soluble protein to a rabbit reticulocyte cell-free system at a concentration of 6.2 microM led to an almost complete inhibition of protein synthesis. The kinetics are unlike those of hemin-controlled inhibitor, a protein that acts at the initiation step. The inhibition appears to be due to an endoribonucleolytic activity of perchloric acid-soluble protein because L-PSP directly affects mRNA template activity and induces disaggregation of the reticulocyte polysomes into 80 S ribosomes, even in the presence of cycloheximide. These effects were observed with authentic as well as recombinant L-PSP. Analysis by thin-layer chromatography of [alpha-32P]UTP-labeled mRNA incubated with the protein showed production of the ribonucleoside 3'-monophosphates Ap, Gp, Up, and Cp, providing direct evidence that the protein is an endoribonuclease. When either 5'- or 3'-32P-labeled 5 S rRNA was the substrate, L-PSP cleaved phosphodiester bonds only in the single-stranded regions of the molecule.

L7 ANSWER 37 OF 103 MEDLINE on STN  
ACCESSION NUMBER: 94024869 MEDLINE Full-text  
DOCUMENT NUMBER: PubMed ID: 8211976  
TITLE: Production of dihydrofolate reductase by an improved continuous flow cell-free translation system using wheat germ extract.  
AUTHOR: Endo Y; Oka T; Ogata K; Natori Y  
CORPORATE SOURCE: Department of Biochemistry, Yamanashi Medical College, Japan.  
SOURCE: The Tokushima journal of experimental medicine, (1993 Jun) Vol. 40, No. 1-2, pp. 13-7.

Journal code: 0417356. ISSN: 0040-8875.  
 PUB. COUNTRY: Japan  
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
 LANGUAGE: English  
 FILE SEGMENT: Priority Journals  
 ENTRY MONTH: 199311  
 ENTRY DATE: Entered STN: 17 Jan 1994  
 Last Updated on STN: 17 Jan 1994  
 Entered Medline: 19 Nov 1993

ED Entered STN: 17 Jan 1994  
 Last Updated on STN: 17 Jan 1994  
 Entered Medline: 19 Nov 1993

AB We have examined the characteristics of protein synthesis in an improved continuous flow cell-free translation system prepared from wheat germ extract with dihydrofolate reductase mRNA as the translated message. Continuous buffer flow and separation of the product from the reaction mixture were accomplished by the use of a modified Amicon ultrafiltration chamber as the reaction vessel. The system worked for 19 hours and produced 1.52 nmol (27.4 micrograms) of enzymatically active dihydrofolate reductase.

L7 ANSWER 38 OF 103 MEDLINE on STN

ACCESSION NUMBER: 94024866 MEDLINE Full-text  
 DOCUMENT NUMBER: PubMed ID: 8211973  
 TITLE: The biosynthesis of a cytotoxic protein, alpha-sarcin, in a mold *Aspergillus giganteus*. I. Synthesis of prepro- and pro-alpha-sarcin in vitro.  
 AUTHOR: Endo Y; Oka T; Tsurugi K; Natori Y  
 CORPORATE SOURCE: Department of Biochemistry, Yamaguchi Medical College, Japan.  
 SOURCE: The Tokushima journal of experimental medicine, (1993 Jun) Vol. 40, No. 1-2, pp. 1-6.

Journal code: 0417356. ISSN: 0040-8875.  
 PUB. COUNTRY: Japan  
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
 LANGUAGE: English  
 FILE SEGMENT: Priority Journals  
 ENTRY MONTH: 199311  
 ENTRY DATE: Entered STN: 17 Jan 1994  
 Last Updated on STN: 17 Jan 1994  
 Entered Medline: 19 Nov 1993

ED Entered STN: 17 Jan 1994  
 Last Updated on STN: 17 Jan 1994  
 Entered Medline: 19 Nov 1993

AB The biosynthesis of alpha-sarcin, a ribosome inactivating protein (16.9 KDa) was studied in a mold *Aspergillus giganteus*. The fungus begins to secrete alpha-sarcin after reaching a stationary phase around 50 h of culture. The synthesis of alpha-sarcin was shown to be induced at the transcriptional level since the mRNA level of alpha-sarcin, titrated by immuno-precipitation with anti-alpha-sarcin antibodies of translation products in wheat germ cell-free system, was increased synchronously with the production of the protein. The immuno-precipitates specific for alpha-sarcin contained two species of proteins of 22.5 and 18.5 KDa. The former was localized in the supernatant and the latter was segregated in the microsomes of the wheat germ system. The 22.5 KDa protein was thought to be the primary product of alpha-sarcin, although N-terminal methionine was removed, because it was the only product when the microsomes were solubilized by Triton X-100 prior to translation in the cell-free system. These results indicate that alpha-sarcin is synthesized as 22.5 KDa prepro-alpha-sarcin and is processed cotranslationally into 18.5 KDa pro-alpha-sarcin in endoplasmic reticulum as usual secretory proteins.

L7 ANSWER 39 OF 103 MEDLINE on STN

ACCESSION NUMBER: 93001007 MEDLINE Full-text

DOCUMENT NUMBER: PubMed ID: 1368801

TITLE: Production of an enzymatic active protein using a continuous flow cell-free translation system.

AUTHOR: Endo Y; Otsuzuki S; Ito K; Miura K

CORPORATE SOURCE: Department of Biochemistry, Yamanashi Medical College, Japan.

SOURCE: Journal of biotechnology, (1992 Sep) Vol. 25, No. 3, pp. 221-30.

Journal code: 8411927. ISSN: 0168-1656.

PUB. COUNTRY: Netherlands

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English

FILE SEGMENT: Biotechnology

ENTRY MONTH: 199211

ENTRY DATE: Entered STN: 9 Aug 1995

Last Updated on STN: 9 Aug 1995

Entered Medline: 3 Nov 1992

ED Entered STN: 9 Aug 1995

Last Updated on STN: 9 Aug 1995

Entered Medline: 3 Nov 1992

AB We have examined the characteristics of protein synthesis in an improved continuous flow cell-free translation system prepared from wheat germ extract with dihydrofolate reductase (dhfr) mRNA as the translated message. Continuous buffer flow and separation of product from the reaction mixture were accomplished by the use of a modified Amicon ultrafiltration chamber as reaction vessel. The system produced protein for more than 20 h, and the product had an activity of dhfr comparable to that of authentic enzyme from E. coli. Analysis of RNA recovered from the filtrate supports the notion that a functionally active protein-synthesizing machinery is superorganized in a dynamic complex.

L7 ANSWER 40 OF 103 MEDLINE on STN

ACCESSION NUMBER: 88198077 MEDLINE Full-text

DOCUMENT NUMBER: PubMed ID: 2452154

TITLE: Characterization of a novel acidic protein of 38 kDa, A0, in yeast ribosomes which immunologically cross-reacts with the 13 kDa acidic ribosomal proteins, A1/A2.

AUTHOR: Mitsui K; Motizuki M; Endo Y; Yokota S; Tsurugi K

CORPORATE SOURCE: Department of Biochemistry, Yamanashi Medical College.

SOURCE: Journal of biochemistry, (1987 Dec) Vol. 102, No. 6, pp. 1565-70.

Journal code: 0376600. ISSN: 0021-924X.

PUB. COUNTRY: Japan

DOCUMENT TYPE: (COMPARATIVE STUDY)

Journal; Article; (JOURNAL ARTICLE)

(RESEARCH SUPPORT, NON-U.S. GOV'T)

LANGUAGE: English

FILE SEGMENT: Priority Journals

ENTRY MONTH: 198806

ENTRY DATE: Entered STN: 8 Mar 1990

Last Updated on STN: 29 Jan 1996

Entered Medline: 2 Jun 1988

ED Entered STN: 8 Mar 1990

Last Updated on STN: 29 Jan 1996

Entered Medline: 2 Jun 1988

AB A new ribosomal protein of 38 kDa, named A0, was detected in yeast ribosomes on immunoblotting. The antibody used here was that against A1/A2, 13 kDa acidic ribosomal proteins which cross-reacted with A0. Although A0 and A1/A2

share common antigenic determinants, they differ in the following biochemical properties. While A1/A2 could be extracted from ribosomes with ethanol and ammonium sulfate, A0 could not. A0 gave two protein spots in a less acidic region than for A1/A2 on two-dimensional gel electrophoresis. The heterogeneity observed for A0 was ascribable to phosphorylation because one spot disappeared after treatment of the ribosomes with phosphatase. The syntheses of A0 and A1/A2 are directed by different mRNA species, as judged with a cell-free translation system, ruling out the possibility that A0 is a precursor of A1/A2. Although a mammalian ribosomal protein equivalent to A0 has been shown to be associated with 13 kDa acidic proteins in the cytoplasm, essentially no A0 was detected on immunoblotting in the yeast cytosol, while a small but detectable amount of A1/A2 was present. The possibility that A0 is a eukaryotic equivalent of L10 of *Escherichia coli* is discussed.

L7 ANSWER 41 OF 103 MEDLINE on STN

ACCESSION NUMBER: 82239263 MEDLINE Full-text

DOCUMENT NUMBER: PubMed ID: 7096300

TITLE: A cell-free system from ethionine-treated rat liver active in initiation of protein synthesis.

AUTHOR: Hase M; Endo Y; Natori Y

SOURCE: Journal of biochemistry, (1982 May) Vol. 91, No. 5, pp. 1457-65.

Journal code: 0376600. ISSN: 0021-924X.

PUB. COUNTRY: Japan

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English

FILE SEGMENT: Priority Journals

ENTRY MONTH: 198209

ENTRY DATE: Entered STN: 17 Mar 1990

Last Updated on STN: 17 Mar 1990

Entered Medline: 17 Sep 1982

ED Entered STN: 17 Mar 1990

Last Updated on STN: 17 Mar 1990

Entered Medline: 17 Sep 1982

AB A cell-free protein-synthesizing system active in initiation of translation of both endogenous mRNA and exogenous mRNA has been obtained from postmitochondrial supernatant (S-12) of the liver of ethionine-treated rats by adding reticulocyte ribosomal extract as a source of initiation factor. Formation of polysomes in the course of protein synthesis *in vitro* has also been demonstrated. Homogenization of the liver in the presence of 50 microM hemin stabilizes the initiation activity of S-12 fraction, which otherwise decays rapidly even at 0 degrees C. The mechanism of inhibition of protein synthesis by ethionine is discussed in view of these results.

L7 ANSWER 42 OF 103 MEDLINE on STN

ACCESSION NUMBER: 79063686 MEDLINE Full-text

DOCUMENT NUMBER: PubMed ID: 82483

TITLE: Protein synthesis by rat transplantable yolk sac tumor and its relation to the cytosol levels of translatable messenger RNA's.

AUTHOR: Kaneko Y; Endo Y; Oda T

SOURCE: Cancer research, (1978 Dec) Vol. 38, No. 12, pp. 4728-33.

Journal code: 2984705R. ISSN: 0008-5472.

PUB. COUNTRY: United States

DOCUMENT TYPE: (COMPARATIVE STUDY)

(IN VITRO)

Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English

FILE SEGMENT: Priority Journals

ENTRY MONTH: 197902  
ENTRY DATE: Entered STN: 14 Mar 1990  
Last Updated on STN: 14 Mar 1990  
Entered Medline: 12 Feb 1979

ED Entered STN: 14 Mar 1990  
Last Updated on STN: 14 Mar 1990  
Entered Medline: 12 Feb 1979

AB alpha-Fetoprotein (AFP) was shown to be the major secretory protein produced in vitro by normal rat yolk sacs. While not so active, AFP production was also detected in the transplantable tumors derived from normal yolk sacs. The major secretory protein synthesized by the tumor cells had a molecular weight of 40,000 and was reactive with an anti-rat albumin antibody. The functional messenger RNA's coding for these proteins were quantitated by translation in a cell-free system derived from wheat germ followed by specific immunoprecipitation of the newly synthesized peptides. The overall template activity of the RNA prepared from the normal yolk sacs and yolk sac tumor cells was virtually identical. The cytosol RNA prepared from the normal yolk sacs was approximately 12 times more active than that from the tumor cells in directing the synthesis of AFP. The presence of the cytosol RNA prepared from the tumor cells was required for the synthesis of proteins immunoprecipitable with the antialbumin antibody. These results suggest that the changes in AFP and albumin synthesis can be accounted for by a corresponding change in the levels of functional messenger RNA's coding for these proteins.

L7 ANSWER 43 OF 103 BIOSIS COPYRIGHT (c) 2008 The Thomson Corporation on STN  
ACCESSION NUMBER: 2008:243702 BIOSIS Full-text  
DOCUMENT NUMBER: PREV200800243303

TITLE: Expression of malaria vaccine candidates using a wheat germ cell-free protein synthesis system without codon optimisation.

AUTHOR(S): Tsuboi, Takafumi [Reprint Author]; Takeo, Satoru; Iriko, Hideyuki; Jin, Ling; Tsuchimochi, Masateru; Matsuda, Shusaku; Han, Eun-Taek; Otsuki, Hitoshi; Kaneko, Osamu; Sattabongkot, Jetsumon; Udomsangpetch, Rachanee; Sawasaki,

Tatsuya; Torii, Motomi; Endo, Yaeta  
CORPORATE SOURCE: Ehime Univ, Cell Free Sci and Technol Res Ctr, Matsuyama, Ehime, Japan

SOURCE: International Journal for Parasitology, (JAN 2008) Vol. 38, No. Suppl. 1, pp. S77.

Meeting Info.: 3rd Molecular Approaches to Malaria Meeting (MAM 2008). Lorne, AUSTRALIA. February 03 -07, 2008. BioMalPar; Boehringer Ingelheim Foods; Burroughs Wellcome Fund; Fdn Natl Inst Hlth; PATH Malaria Vaccine Initiative;

Walter & Eliza Hall Inst Med Res; Wellcome Trust; ARC/NHMRC Net Parasitol; Australian Soc Biochem & Molecular Biol; Lorne Protein Conf; GlaxoSmithKline.

CODEN: IJPBYT. ISSN: 0020-7519.

DOCUMENT TYPE: Conference; (Meeting)  
Conference; (Meeting Poster)

LANGUAGE: English

ENTRY DATE: Entered STN: 2 Apr 2008  
Last Updated on STN: 2 Apr 2008

ED Entered STN: 2 Apr 2008  
Last Updated on STN: 2 Apr 2008

L7 ANSWER 44 OF 103 BIOSIS COPYRIGHT (c) 2008 The Thomson Corporation on STN  
ACCESSION NUMBER: 2007:605234 BIOSIS Full-text  
DOCUMENT NUMBER: PREV200700606893

TITLE: Methods for high-throughput materialization of genetic information based on wheat germ cell-free expression system.

AUTHOR(S): Sawasaki, Tatsuya [Reprint Author]; Morishita, Ryo; Gouda, Mudeppa D.; Endo, Yaeta  
CORPORATE SOURCE: Ehime Univ, Cell Free Sci and Technol Res Ctr, Matsuyama, Ehime 790, Japan  
SOURCE: Grandi, G [Editor]. Methods in Molecular Biology, (2007) pp. 95-106. Methods in Molecular Biology.  
Publisher: HUMANA PRESS INC, 999 RIVERVIEW DR, STE 208, TOTOWA, NJ 07512-1165 USA. Series: METHODS IN MOLECULAR BIOLOGY.  
ISSN: 1064-3745. ISBN: 978-1-58829-558-3(H).  
DOCUMENT TYPE: Book; (Book Chapter)  
LANGUAGE: English  
ENTRY DATE: Entered STN: 6 Dec 2007  
Last Updated on STN: 6 Dec 2007

ED Entered STN: 6 Dec 2007  
Last Updated on STN: 6 Dec 2007

AB Among the cell-free protein synthesis systems, the wheat germ-based translation system has significant advantages for the high-throughput production of eukaryotic multidomain proteins in folded state. Here, we describe protocols for this cell-free expression system.

L7 ANSWER 45 OF 103 BIOSIS COPYRIGHT (c) 2008 The Thomson Corporation on STN

ACCESSION NUMBER: 2007:442841 BIOSIS [Full-text](#)  
DOCUMENT NUMBER: PREV200700439848  
TITLE: Preparation containing cell extracts for cell-free protein synthesis and means for synthesizing protein using the preparation.  
AUTHOR(S): Anonymous; Endo, Yaeta [Inventor]; Nishikawa, Shigemichi [Inventor]  
CORPORATE SOURCE: Matsuyama, Japan  
ASSIGNEE: CellFree Sciences Co Ltd  
PATENT INFORMATION: US 0735382 20070626  
SOURCE: Official Gazette of the United States Patent and Trademark Office Patents, (JUN 26 2007)  
CODEN: OGUPE7. ISSN: 0098-1133.

DOCUMENT TYPE: Patent  
LANGUAGE: English  
ENTRY DATE: Entered STN: 15 Aug 2007  
Last Updated on STN: 15 Aug 2007

ED Entered STN: 15 Aug 2007

Last Updated on STN: 15 Aug 2007

AB Disclosed are a preparation containing cell extracts for cell-free protein synthesis, prepared by excluding from a living organism a system, participating to inhibiting of self protein synthesis reaction, an apparatus for cell-free protein synthesis reaction equipped with a reaction tank for cell-free protein synthesis, and a kit for use therefor; the preparation can be stored at room temperature and prepared as a preparation in a state where biological functions of the cell extracts are maintained, and further, disclosed is methods for cell-free protein synthesis comprising providing cell extracts from which an inhibitor for self protein synthesis reaction is substantially excluded, having introduced therein treatment selected from supplement, storage, exchange or discharge with respect to an element selected from at least mRNA serving as a template for synthesis reaction, an energy reproduction system enzyme, a substrate, and an energy source.

L7 ANSWER 46 OF 103 BIOSIS COPYRIGHT (c) 2008 The Thomson Corporation on STN

ACCESSION NUMBER: 2006:669281 BIOSIS [Full-text](#)  
DOCUMENT NUMBER: PREV200600682572  
TITLE: General means of labeling protein by using wheat embryo cell-free protein synthesis system.

AUTHOR(S): Anonymous; Endo, Yaeta [Inventor]; Kumar, Penmetcha [Inventor];  
Nishikawa, Shigemichi [Inventor]  
CORPORATE SOURCE: Matsuyama, Japan  
ASSIGNEE: CellFree Sciences Co Ltd  
PATENT INFORMATION: US 07074595 20060711  
SOURCE: Official Gazette of the United States Patent and Trademark  
Office Patents, (JUL 11 2006)  
CODEN: OGPU7. ISSN: 0098-1133.  
DOCUMENT TYPE: Patent  
LANGUAGE: English  
ENTRY DATE: Entered STN: 6 Dec 2006  
Last Updated on STN: 6 Dec 2006

ED Entered STN: 6 Dec 2006

Last Updated on STN: 6 Dec 2006

AB Utilizing a wheat embryo cell-free protein synthesis system, there are provided a process for the production of selenomethionine-labeled protein, characterized in that, methionine in a wheat embryo extract for a cell-free protein synthesis obtained by a complete removal of endosperm contaminated is changed to selenomethionine and a cell-free protein synthesis is carried out using a reaction solution composition for protein synthesis containing selenomethionine instead of methionine under a batch condition or a dialysis condition and also the said protein produced as such. There are further provided a process for the production of heavy hydrogen-labeled protein using the same means and also the said protein produced as such.

L7 ANSWER 47 OF 103 BIOSIS COPYRIGHT (c) 2008 The Thomson Corporation on STN

ACCESSION NUMBER: 2006:575286 BIOSIS Full-text

DOCUMENT NUMBER: PREV200600571389

TITLE: Covalent circularization of exogenous RNA during incubation with a wheat embryo cell extract.

AUTHOR(S): Makino, Shin-ichi; Sawasaki, Tatsuya; Tozawa, Yuzuru; Endo, Yaeta; Takai, Kazuyuki [Reprint Author]

CORPORATE SOURCE: Ehime Univ, Venture Business Lab, 3 Bunkyo Cho, Matsuyama, Ehime 7908577, Japan

takai@eng.ehime-u.ac.jp

SOURCE: Biochemical and Biophysical Research Communications, (SEP 8 2006) Vol. 347, No. 4, pp. 1080-1087.

CODEN: BBRCA9. ISSN: 0006-291X.

DOCUMENT TYPE: Article

LANGUAGE: English

ENTRY DATE: Entered STN: 1 Nov 2006

Last Updated on STN: 1 Nov 2006

ED Entered STN: 1 Nov 2006

Last Updated on STN: 1 Nov 2006

AB Cell extracts from wheat embryos have been widely used for mRNA-directed protein production. Here, we found that a significant fraction of exogenous linear RNAs are circularized in a wheat embryo extract. The circularization was seen only in uncapped RNAs. The amount of the circular species reached around 1% of the initial RNA and increased along with an increase in the initial concentration more than proportionally. The circular RNAs were stable but unable to be translated in the extract. The circularization was competitively inhibited in the presence of a known substrate of a wheat embryo RNA ligase. Thus, we cloned the RNA ligase cDNAs. Three isoform sequences were homologous to the other plant RNA ligases. An addition of a cell-free synthesized wheat RNA ligase abolished the inhibition, which indicates a participation of its activity in the circularization. A possible role in RNA metabolism, RNA silencing in particular, is discussed. (c) 2006 Elsevier Inc. All rights reserved.



L7 ANSWER 48 OF 103 BIOSIS COPYRIGHT (c) 2008 The Thomson Corporation on STN  
ACCESSION NUMBER: 2006:212186 BIOSIS Full-text  
DOCUMENT NUMBER: PREV200600215350  
TITLE: Preparation containing cell extracts for synthesizing cell-free  
protein and means for synthesizing cell-free protein.  
AUTHOR(S): Endo, Yaeta [Inventor]; Nishikawa, Shigemichi [Inventor]  
CORPORATE SOURCE: Matsuyama, Japan  
ASSIGNEE: CellFree Sciences Co., Ltd.  
PATENT INFORMATION: US 06905843 20050614  
SOURCE: Official Gazette of the United States Patent and Trademark  
Office Patents, (JUN 14 2005)  
CODEN: OGUPE7. ISSN: 0098-1133.  
DOCUMENT TYPE: Patent  
LANGUAGE: English  
ENTRY DATE: Entered STN: 29 Mar 2006  
Last Updated on STN: 29 Mar 2006

ED Entered STN: 29 Mar 2006

Last Updated on STN: 29 Mar 2006

AB Disclosed are a preparation containing cell extracts for cell-free protein  
synthesis, prepared by excluding from a living organism a system,  
participating to inhibiting of self protein synthesis reaction, an apparatus  
for cell-free protein synthesis reaction equipped with a reaction tank for  
cell-free protein synthesis, and a kit for use thereof; the preparation can  
be stored at room temperature and prepared as a preparation in a state where  
biological functions of the cell extracts are maintained and further,  
disclosed is means for cell-free protein synthesis comprising cell extracts  
from which an inhibitor for self protein synthesis reaction is substantially  
excluded, having introduced therein treatment selected from supplement,  
storage, exchange or discharge with respect to an element selected from at  
least mRNA serving as a template for synthesis reaction, an energy  
reproduction system enzyme, a substrate, and an energy source.

L7 ANSWER 49 OF 103 BIOSIS COPYRIGHT (c) 2008 The Thomson Corporation on STN  
ACCESSION NUMBER: 2006:133738 BIOSIS Full-text  
DOCUMENT NUMBER: PREV200600143348  
TITLE: Methods of synthesizing cell-free protein.  
AUTHOR(S): Endo, Yaeta [Inventor]; Sawasaki, Tatsuya [Inventor];  
Ogasawara, Tomio [Inventor]  
CORPORATE SOURCE: Ehime 791-8016, Japan  
ASSIGNEE: Yaeta Endo  
PATENT INFORMATION: US 06869774 20050322  
SOURCE: Official Gazette of the United States Patent and Trademark  
Office Patents, (MAR 22 2005)  
CODEN: OGUPE7. ISSN: 0098-1133.

DOCUMENT TYPE: Patent  
LANGUAGE: English  
ENTRY DATE: Entered STN: 22 Feb 2006  
Last Updated on STN: 22 Feb 2006

ED Entered STN: 22 Feb 2006

Last Updated on STN: 22 Feb 2006

AB One embodiment of the present invention is a diffusion continuous batch cell-  
free protein-synthesis method characterized simultaneously by continuously  
supplying substrate and energy source molecules in the supply phase to the  
reaction phase by the free diffusion via interface between both phases and by  
transferring by-products formed in the reaction phase by enhancing the  
efficiency of the synthesis reaction by prolonging the reaction lifetime by  
directly contacting a synthesis reaction mixture (reaction phase) containing a  
biological extract with a substrate- and energy source-supplying solution

(supply phase) without using barrier such as semi-permeable membrane or ultrafiltration membrane in a general cell-free protein-synthesis reaction means. Another embodiment of the present invention is a dilution batch cell-free protein synthesis method characterized by enhancing the efficiency of the protein synthesis by prolonging the reaction lifetime by adding a diluting solution to the reaction mixture after pre-incubating the reaction mixture in a cell-free protein-synthesis reaction means using a wheat-embryo extract. Another embodiment of the present invention is a method characterized by enhancing the efficiency of the synthesis reaction simultaneously by re-supplying substrate and energy sources necessary for the protein synthesis (e.g., amino acids, ATP, GTP, creatine phosphate) to the reaction mixture using a gel filtration column and/or semipermeable membrane and by discontinuously removing by-products formed during the reaction after the synthesis reaction stops in the batch cell-free protein synthesis method.

L7 ANSWER 50 OF 103 BIOSIS COPYRIGHT (c) 2008 The Thomson Corporation on STN

ACCESSION NUMBER: 2002:158200 BIOSIS Full-text

DOCUMENT NUMBER: PREV200200158200

TITLE: Recent advances in the cell-free protein synthesis system.

AUTHOR(S): Gawasaki, Tatsuya [Reprint author]; Endo, Yaeta [Reprint author]

CORPORATE SOURCE: Applied Chemistry, Ehime University, Matsuyama, Japan

SOURCE: Molecular Biology of the Cell, (Nov, 2001) Vol. 12, No.

Supplement, pp. 392a. print.

Meeting Info.: 41st Annual Meeting of the American Society for Cell Biology. Washington DC, USA. December 08-12, 2001. American Society for Cell Biology.

DOCUMENT TYPE: CODEN: MBCEEV. ISSN: 1059-1524.

Conference; (Meeting)

Conference; Abstract; (Meeting Abstract)

LANGUAGE: English

ENTRY DATE: Entered STN: 21 Feb 2002

Last Updated on STN: 26 Feb 2002

ED Entered STN: 21 Feb 2002

Last Updated on STN: 26 Feb 2002

L7 ANSWER 51 OF 103 BIOSIS COPYRIGHT (c) 2008 The Thomson Corporation on STN

ACCESSION NUMBER: 1979:230748 BIOSIS Full-text

DOCUMENT NUMBER: PREV1979968033252; BA68:33252

TITLE: DIRECT ASSOCIATION OF MESSENGER RNA CONTAINING RIBO NUCLEO

PROTEIN PARTICLES WITH MEMBRANES OF THE ENDOPLASMIC RETICULUM IN ETHIONINE TREATED RAT LIVER.

AUTHOR(S): ENDO Y [Reprint author]; NATORI Y

CORPORATE SOURCE: DEP NUTR CHEM, TOKUSHIMA UNIV SCH MED, TOKUSHIMA 770, JPN

SOURCE: Biochimica et Biophysica Acta, (1979) Vol. 562, No. 2, pp. 281-291.

CODEN: BBACAQ. ISSN: 0006-3002.

DOCUMENT TYPE: Article

FILE SEGMENT: BA

LANGUAGE: ENGLISH

AB The administration of ethionine to female rats causes breakdown of hepatic polysomes. The fate of the mRNA molecules after polysome breakdown was investigated by measuring the amount of poly(A)-containing mRNA in membranous and non-membranous fractions obtained from the cytoplasm of ethionine-treated rat liver. The amount of poly(A)-containing mRNA in the membrane fraction of ethionine-treated liver was the same as that of normal liver. When poly(A)-containing mRNAs from various fractions were translated in a wheat germ system and the products were isolated by immunoprecipitation, the albumin-specific mRNA was found exclusively in the membrane fraction of both normal and

ethionine-treated livers. The membrane-bound mRNA in ethionine-treated liver, selectively labeled with [<sup>14</sup>C]orotate, was banded in CsCl gradient centrifugation at 1.42 g/ml which corresponds to the previously reported mRNA-containing ribonucleoprotein particles. Even after the polysome disaggregation by ethionine, most of the mRNA of membrane-bound polysomes apparently remains attached to the endoplasmic reticulum membranes independently of ribosomes and the nascent polypeptide chains.

L7 ANSWER 52 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN  
ACCESSION NUMBER: 2008:527505 CAPLUS Full-text  
TITLE: A set of ligation-independent in vitro translation vectors for eukaryotic protein production  
AUTHOR(S): Bardoczy, Viola; Geczi, Viktoria; Sawasaki, Tatsuya; Endo, Yaeta; Meszaros, Tamas  
CORPORATE SOURCE: Department of Applied Biotechnology and Food Science, Budapest University of Technology and Economics, Budapest, Hung.  
SOURCE: BMC Biotechnology (2008), 8, No pp. given  
CODEN: BBMIE6; ISSN: 1472-6750  
URL: [http://www.biomedcentral.com/content/pdf/1472-6750-8-](http://www.biomedcentral.com/content/pdf/1472-6750-8-32.pdf)

32.pdf

PUBLISHER: BioMed Central Ltd.  
DOCUMENT TYPE: Journal; (online computer file)  
LANGUAGE: English  
ED Entered STN: 01 May 2008

AB Background: The last decade has brought the renaissance of protein studies and accelerated the development of high-throughput methods in all aspects of proteomics. Presently, most protein synthesis systems exploit the capacity of living cells to translate proteins, but their application is limited by several factors. A more flexible alternative protein production method is the cell-free in vitro protein translation. Currently available in vitro translation systems are suitable for high-throughput robotic protein production, fulfilling the requirements of proteomics studies. Wheat germ extract based in vitro translation system is likely the most promising method, since numerous eukaryotic proteins can be cost-efficiently synthesized in their native folded form. Although currently available vectors for wheat embryo in vitro translation systems ensure high productivity, they do not meet the requirements of state-of-the-art proteomics. Target genes have to be inserted using restriction endonucleases and the plasmids do not encode cleavable affinity purification tags. Results: We designed four ligation independent cloning (LIC) vectors for wheat germ extract based in vitro protein translation. In these constructs, the RNA transcription is driven by T7 or SP6 phage polymerase and two TEV protease cleavable affinity tags can be added to aid protein purifn. To evaluate our improved vectors, a plant mitogen activated protein kinase was cloned in all four constructs. Purifn. of this eukaryotic protein kinase demonstrated that all constructs functioned as intended: insertion of PCR fragment by LIC worked efficiently, affinity purifn. of translated proteins by GST-Sepharose or MagneHis particles resulted in high purity kinase, and the affinity tags could efficiently be removed under different reaction conditions. Furthermore, high in vitro kinase activity testified of proper folding of the purified protein. Conclusion: Four newly designed in vitro translation vectors have been constructed which allow fast and parallel cloning and protein purifn., thus representing useful mol. tools for high-throughput prodn. of eukaryotic proteins.

L7 ANSWER 53 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN  
ACCESSION NUMBER: 2008:420693 CAPLUS Full-text  
TITLE: Hetero subunits assembly study of RNA modification enzyme by wheat germ cell-free translation system

AUTHOR(S): Matsumoto, Keisuke; Abe, Masato; Takano, Yoshitaka;  
Takayanagi, Naoyuki; Endo, Yaeta; Hori, Hiroyuki  
CORPORATE SOURCE: Graduate School of Materials Science and Biotechnology,  
Ehime University, Japan  
SOURCE: IEEE International Symposium on Micro-NanoMechatronics and  
Human Science, Nagoya, Japan, Nov. 6-8, 2006 (2006), 328-333. Institute of  
Electrical and Electronics Engineers: New York, N. Y.  
CODEN: 69KOAR; ISBN: 1-4244-0717-6  
DOCUMENT TYPE: Conference  
LANGUAGE: English

ED Entered STN: 04 Apr 2008

AB In the living cells, many kinds of modified nucleosides exist in various RNA species. These modified nucleosides are generated by specific RNA modification enzymes. In almost cases, RNA modification enzyme is composed by one single subunit or homo-subunits. However, recent studies have revealed that eukaryote tRNA (m7G46) methyltransferases are exceptionally constituted by hetero-subunits (Trm8/Trm82 in yeast; METTL1/WDRA in human). This enzyme catalyzes the methyl-transfer from S-adenosyl-L-methionine to the N7 atom of the semi-conserved guanosine at position 46 in the extra-loop of tRNA. To clarify the functions of two subunits, we employed wheat germ cell-free translation system. When the Trm8 or Trm82 subunit alone was synthesized, methyl-transfer activity was not detectable. In contrast, when both Trm8 and Trm82 subunits were synthesized together, tRNA methyltransferase activity was clearly detected. Furthermore, we mixed two subunits after the synthesis, however formation of the active hetero-dimer was not observed. These results demonstrated that the formation of the active Trm8 and Trm82 hetero-dimer requires the subunit-subunit interaction during the protein synthesis.

L7 ANSWER 54 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN

ACCESSION NUMBER: 2008:98117 CAPLUS [Full-text](#)  
DOCUMENT NUMBER: 148:489844  
TITLE: The wheat germ cell-free protein synthesis system  
AUTHOR(S): Sawasaki, Tatsuya; Endo, Yaeta  
CORPORATE SOURCE: Cell-Free Science and Technology Research Center, Ehime University, Matsuyama, 790-8577, Japan  
SOURCE: Cell-Free Protein Synthesis (2008), 111-139. Editor(s): Spirin, Alexander S.; Swartz, James R. Wiley-VCH Verlag GmbH & Co. KGaA: Weinheim, Germany.

CODEN: 69KIQP; ISBN: 978-3-527-31649-6  
DOCUMENT TYPE: Conference  
LANGUAGE: English

ED Entered STN: 25 Jan 2008

AB Among cell-free protein synthesis systems, the wheat germ-based translation system is of special interest for its eukaryotic nature: it has significant advantages for the high-throughput production of eukaryotic multi-domain proteins in the folded state. Here the authors describe how this highly efficient cell-free expression system is built and review its application to today's functional and structural biol.

L7 ANSWER 55 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN

ACCESSION NUMBER: 2007:871114 CAPLUS [Full-text](#)  
DOCUMENT NUMBER: 147:253468  
TITLE: Biotinylated protein preparation method, and detection method using biotinylated protein  
INVENTOR(S): Endo, Yaeta; Sawasaki, Tatsuya; Matsubara, Yuko  
PATENT ASSIGNEE(S): Cellfree Sciences Co., Ltd., Japan  
SOURCE: Jpn. Kokai Tokkyo Koho, 20pp.  
CODEN: JKXXAF

DOCUMENT TYPE: Patent  
 LANGUAGE: Japanese  
 FAMILY ACC. NUM. COUNT: 1  
 PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
JP 2007199047	A	20070809	JP 2006-182785	20060630
US 20070190579	A1	20070816	US 2006-643737	20061221
PRIORITY APPLN. INFO.:			JP 2005-377840	A 20051228
			JP 2006-182785	A 20060630

ED Entered STN: 09 Aug 2007

AB A biotinylated protein preparation method is provided, which enables to prepare a biotin-tagged protein to be used in a detection method (e.g., ALPHA, SPR, FCS, FIDA, ELISA, DELFIA, SPA, FRET, BRET, EFC, FP) for a substance capable of interacting with the protein without requiring a free biotin removal process. In this method for preparing a biotinylated protein, a protein biotinylation is achieved with a biotin derivative of a remarkably lower concentration than the conventional biotinylation operation by carrying out a biotinylation using a biotinylation enzyme and the biotin derivative during or after a protein synthesis using a cell-free protein synthesis system, especially, a wheat germ cell-free protein synthesis system.

L7 ANSWER 56 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN

ACCESSION NUMBER: 2007:867314 CAPLUS [Full-text](#)  
 DOCUMENT NUMBER: 147:184148  
 TITLE: Protein-binding carrier for efficient recovery of protein  
 in cell-free translation  
 INVENTOR(S): Endo, Yaeta  
 PATENT ASSIGNEE(S): Cellfree Sciences Co., Ltd., Japan  
 SOURCE: Jpn. Kokai Tokkyo Koho, 18pp.  
 CODEN: JKXXAF

DOCUMENT TYPE: Patent  
 LANGUAGE: Japanese  
 FAMILY ACC. NUM. COUNT: 1  
 PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
JP 2007195545	A	20070809	JP 2006-350351	20061226
PRIORITY APPLN. INFO.:			JP 2005-372957	A 20051226

ED Entered STN: 09 Aug 2007

AB The protein-binding carrier is selected from specific protein-binding substance-immobilized bead, magnetic bead, ion-exchanger, and affinity resin. The protein binding carrier is useful for prevention of clogging of membrane of protein in com. manufacture of protein by cell-free translation. It is also allowing repeated use of the cell-free translation system. Also, the protein thus obtain retain original structure. Recovery of green fluorescence protein which is tagged with glutathione-S transferase in wheat germ cell-free translation system with glutathione magnetic bead was shown.

L7 ANSWER 57 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN

ACCESSION NUMBER: 2007:838548 CAPLUS [Full-text](#)  
 DOCUMENT NUMBER: 147:210145  
 TITLE: Preparation of antigens from Rubella virus using wheat  
 germ protein synthesis system and their use for evaluation of the risk of  
 congenital rubella syndrome  
 INVENTOR(S): Endo, Yaeta; Tsuboi, Takafumi; Okuyama, Masaaki; Oseto,

Mitsuaki  
 PATENT ASSIGNEE(S): Cellfree Sciences Co., Ltd., Japan  
 SOURCE: Jpn. Kokai Tokkyo Koho, 20pp.  
 CODEN: JKXXAF  
 DOCUMENT TYPE: Patent  
 LANGUAGE: Japanese  
 FAMILY ACC. NUM. COUNT: 1  
 PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
JP 2007191477	A	20070802	JP 2006-346072	20061222
PRIORITY APPLN. INFO.:			JP 2005-370320	A 20051222

ED Entered STN: 02 Aug 2007

AB This invention provides a process of preparation of antigens using wheat germ protein synthesis system in which the endosperm and low mol. protein synthesis inhibitors were eliminated. The proteins were derived from Rubella virus E1 and E2 proteins, and the amino acid sequences of these proteins were disclosed. The proteins were able to cross react to the antibodies in serum of mouse immunized with the antigens. The antigens provided in this invention can be used for preparation of test kit for evaluation of the risk of congenital rubella syndrome in females.

L7 ANSWER 58 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN

ACCESSION NUMBER: 2007:677102 CAPLUS Full-text  
 DOCUMENT NUMBER: 148:373346  
 TITLE: Production of protein for nuclear magnetic resonance study using the wheat germ cell-free system  
 AUTHOR(S): Kohno, Toshiyuki; Endo, Yasta  
 CORPORATE SOURCE: Molecular Structure Research Group, Mitsubishi Kagaku Institute of Life Sciences (MITILS), Tokyo, Japan  
 SOURCE: Methods in Molecular Biology (Totowa, NJ, United States) (2007), 375(In Vitro Transcription and Translation Protocols (2nd Edition)), 257-272

CODEN: MMBIED; ISSN: 1064-3745  
 PUBLISHER: Humana Press Inc.  
 DOCUMENT TYPE: Journal  
 LANGUAGE: English

ED Entered STN: 22 Jun 2007

AB NMR methods have been developed to determine the three-dimensional structures of proteins, to estimate protein folding, and to discover high-affinity ligands for proteins. However, one of the difficulties encountered in the application of such NMR methods to proteins is that the authors should obtain milligram quantities of <sup>15</sup>N and/or <sup>13</sup>C-labeled pure proteins of interest. Here, the authors describe the method to produce proteins for NMR expts. using the improved wheat germ cell-free system, which exhibits several attractive features for high-throughput NMR study of proteins.

REFERENCE COUNT: 12 THERE ARE 12 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 59 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN

ACCESSION NUMBER: 2007:496711 CAPLUS Full-text  
 DOCUMENT NUMBER: 147:270844  
 TITLE: History and use of protein expression system  
 AUTHOR(S): Endo, Yasta  
 CORPORATE SOURCE: Ehime Univ., Japan  
 SOURCE: Biotekunoroji Janaru (2007), 7(2), 230-233  
 CODEN: BJAAA8; ISSN: 1349-7448  
 PUBLISHER: Yodosha

DOCUMENT TYPE: Journal; General Review  
LANGUAGE: Japanese

ED Entered STN: 08 May 2007

AB A review discussed brief history of the development of the technologies for recombinant protein expression systems. Features of the novel protein expression systems recently developed were overviewed. Such expression systems included the system using cold shock vector for improving protein misfolding from TakaraBio Ltd., and the systems to produce proteins with more native characteristics by using "Superwarm" insect cell expression system from Katakura Kogyo Ltd. or by using mammalian cell systems (Invitrogen). Proceeding of the protein expression technol. by using cell free protein synthesis systems was also discussed. Advantages of the cell free system were discussed by comparing E. coli, rabbit reticulocyte and wheat germ lysate systems.

L7 ANSWER 60 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN

ACCESSION NUMBER: 2007:428031 CAPLUS Full-text

DOCUMENT NUMBER: 146:375315

TITLE: Freeze-dried template for cell-free protein synthesis and

laboratory application

INVENTOR(S): Endo, Yaeata; Sawasaki, Tatsuya; Tanaka, Michihiro;

Morishita, Akira; Saeki, Mihoro

PATENT ASSIGNEE(S): Cellfree Sciences Co., Ltd., Japan

SOURCE: Jpn. Kokai Tokkyo Koho, 27pp.

CODEN: JKXXAF

DOCUMENT TYPE: Patent

LANGUAGE: Japanese

FAMILY ACC. NUM. COUNT: 1

PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
-----	-----	-----	-----	-----
JP 2007097438	A	20070419	JP 2005-288613	20050930
PRIORITY APPLN. INFO.:			JP 2005-288613	20050930

ED Entered STN: 19 Apr 2007

AB The freeze-dried template is nucleotide sequence for visible proteins selected from green fluorescent protein, blue fluorescent protein, etc. The RNA polymerase and phosphotransferase such as creatine kinase used in the cell-free protein synthesis do not contain animal and microbial contaminants. With proper solns., the freeze-dried template-containing cell-free protein synthesis system may be easily used, and protein synthesis monitored by the presence of visible proteins. Also, the cell-free protein synthesis system such as wheat germ extract does not use expansive RNase inhibitors.

L7 ANSWER 61 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN

ACCESSION NUMBER: 2007:391722 CAPLUS Full-text

DOCUMENT NUMBER: 146:498906

TITLE: Wheat germ cell-free protein synthesis system: an application to malaria vaccine research

AUTHOR(S): Tsuboi, Takafumi; Takeo, Satoru; Iriko, Hideyuki; Kaneko, Osamu; Torii, Motomi; Endo, Yaeata

CORPORATE SOURCE: Cell-Free Science and Technology Research Center, Ehime University, Japan

SOURCE: Ehime Igaku (2007), 26(1), 8-11

CODEN: EHIGEL; ISSN: 0286-3677

PUBLISHER: Ehime Igakkai

DOCUMENT TYPE: Journal; General Review

LANGUAGE: Japanese

ED Entered STN: 09 Apr 2007

AB A review discusses application of wheat germ cell-free protein system in identification of antigens for malaria vaccine.

L7 ANSWER 62 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN

ACCESSION NUMBER: 2007:185804 CAPLUS [Full-text](#)

DOCUMENT NUMBER: 146:246573

TITLE: Cell-free protein production and its application to post-genomic biotechnology

AUTHOR(S): Endo, Yaeata

CORPORATE SOURCE: Cell-Free Sci. Technol. Res. Cent., Ehime University, Matsuyama, 790-8577, Japan

SOURCE: Baioasaiensu to Indasutori (2007), 65(1), 11-17

CODEN: BIDSE6; ISSN: 0914-8981

PUBLISHER: Baiindasutori Kyokai

DOCUMENT TYPE: Journal; General Review

LANGUAGE: Japanese

ED Entered STN: 20 Feb 2007

AB A review on (1) development of highly effective cell-free protein synthesis system using wheat embryos, (2) improvement of the system by the optimization of the mRNA UTRs, expression vectors, PCR, and translation conditions, (3) development of automated protein synthesis apps., and (4) application of wheat-embryo cell-free protein synthesis system.

L7 ANSWER 63 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN

ACCESSION NUMBER: 2007:49814 CAPLUS [Full-text](#)

DOCUMENT NUMBER: 146:396998

TITLE: Cell-free production of functional Plasmodium falciparum dihydrofolate reductase-thymidylate synthase

AUTHOR(S): Mudeppa, Devaraja G.; Pang, Cullen K. T.; Tsuboi, Takafumi; Endo, Yaeata; Buckner, Fredrick S.; Varani, Gabriele; Rathod, Pradipsinh K.

CORPORATE SOURCE: Department of Chemistry, University of Washington, Seattle, WA, 98195-1700, USA

SOURCE: Molecular & Biochemical Parasitology (2007), 151(2), 216-219

CODEN: MBIPDP; ISSN: 0166-6851

PUBLISHER: Elsevier Ltd.

DOCUMENT TYPE: Journal

LANGUAGE: English

ED Entered STN: 16 Jan 2007

AB Expression of Plasmodium falciparum dihydrofolate reductase-thymidylate synthase (PfDHFR-TS) in a cell-free system is described. Using a SP6 polymerase promoter, the PfDHFR-TS was expressed in the wheat germ system for protein expression. The expressed PfDHFR-TS was purified by dialysis based protocol and the purified protein was characterized. The results show that the amount of the PfDHFR-TS produced in this system was about 100µg of soluble product per mL of the translation reaction thus making this cell free system superior to other systems. The activity of the PfDHFR-TS was determined to be 30nmol/min per mL of the wheat germ extract, and based on the activity of 120/min, this translates into the amts. produced by 4500 mL of the P.falciparum culture. Enzyme kinetics for PfDHFR-TS was also studied.

REFERENCE COUNT: 17 THERE ARE 17 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 64 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN

ACCESSION NUMBER: 2007:14862 CAPLUS [Full-text](#)

DOCUMENT NUMBER: 146:95367



TITLE: Novel use of DNA-binding proteins as fusion protein tags  
 INVENTOR(S): Endo, Yaeta; Sawasaki, Tatsuya; Kamura, Nami; Matsubara, Yuko  
 PATENT ASSIGNEE(S): Cellfree Sciences Co., Ltd., Japan  
 SOURCE: PCT Int. Appl., 51pp.  
 CODEN: PIXXD2  
 DOCUMENT TYPE: Patent  
 LANGUAGE: Japanese  
 FAMILY ACC. NUM. COUNT: 1  
 PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2007000972	A1	20070104	WO 2006-JP312715	20060626
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BW, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, HN, HU, ID, IL, IN, IS, JP, KE, KG, KM, KN, KP, KR, KZ, LA, LC, LK, LR, LS, LT, LU, LV, LY, MA, MD, MG, MK, MN, MW, MX, MZ, NA, NG, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RS, RU, SC, SD, SE, SG, SK, SL, SM, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, ZA, ZM, ZW RW: AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IS, IT, LT, LU, LV, MC, NL, PL, PT, RO, SE, SI, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG, BW, GH, GM, KE, LS, MW, MZ, NA, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM PRIORITY APPLN. INFO.: JP 2005-187468 A 20050627 JP 2005-286956 A 20050930				

ED Entered STN: 05 Jan 2007

AB Disclosed is a novel use of DNA-binding proteins as fusion protein tags for efficient labeling, detection or purification of the target proteins. DNA-binding domains of the binding proteins, more specifically transcription factors or DNA-binding nuclear receptors are used as tags. Three amino acid sequences that specify the DNA-binding proteins are claimed. Tag-moieties is immobilized on carriers by using the interaction of avidin/biotin, maltose-binding protein/maltose, G-protein/guanine nucleotide, DNA-binding protein/DNA, antigen (epitope)/antibody, calmodulin-binding peptide/calmodulin, ATP-binding protein/ATP, or estradiol receptor/estradiol. The fusion protein containing the DNA-binding domain can be captured and purified by using affinity matrixes with DNAs containing corresponding specific sequences. Tags have amino acid sequences sensitive to endopeptidases for proteolysis to recover the target protein. The tagged fusion proteins are designed to be prepared in the cell free protein synthesis system using wheat germ lysate. The tagged fusion proteins are expressed in individual microwells and they are immobilized on carrier matrixes. Microwells containing one or two fusion proteins form protein microarrays (protein chips).

REFERENCE COUNT: 4 THERE ARE 4 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 65 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN

ACCESSION NUMBER: 2006:494180 CAPLUS Full-text

DOCUMENT NUMBER: 145:6561

TITLE: Cell-free protein synthesis systems for detecting antibody and screening specific protein or antigen

INVENTOR(S): Endo, Yaeta; Sawasaki, Tatsuya; Ogasawara, Tomio;

Tsuchimochi, Masateru; Matsubara, Yuko

PATENT ASSIGNEE(S): Cellfree Sciences Co., Ltd., Japan

SOURCE: PCT Int. Appl., 27 pp.

CODEN: PIXXD2

DOCUMENT TYPE: Patent

LANGUAGE: Japanese

FAMILY ACC. NUM. COUNT: 1  
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2006054683	A1	20060526	WO 2005-JP21216	20051118
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BW, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KM, KN, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, LY, MA, MD, MG, MK, MN, MW, MX, MZ, NA, NG, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, SM, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW RW: AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IS, IT, LI, LU, LV, MC, NL, PL, PT, RO, SE, SI, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG, BW, GH, GM, KE, LS, MW, MZ, NA, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM JP 2008035701 A 20080221 JP 2004-335392 20041119 PRIORITY APPLN. INFO.: JP 2004-335392 A 20041119				

ED Entered STN: 26 May 2006

AB It is intended to provide a system whereby an antibody in a specimen can be surely and quickly detected. To achieve this object, various attempts have been made to examine whether or not the cell-free protein synthesis method is applicable to this assay system. As an important factor for achieving the above object, various attempts have been made to improve the expression manner of a specific protein. As a result, it is found out that a target antibody in a specimen can be assayed by preparing a specific protein as a fused protein by the cell-free protein synthesis method and contacting this unpurified fused protein with the specimen.

REFERENCE COUNT: 15 THERE ARE 15 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 66 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN  
 ACCESSION NUMBER: 2006:471739 CAPLUS Full-text  
 DOCUMENT NUMBER: 144:449469  
 TITLE: Cell-free protein synthesis process and apparatus  
 INVENTOR(S): Endo, Yaeta; Sawasaki, Tatsuya; Ogasawara, Tomio;  
 Morishita, Ryo; Saeki, Mihoro  
 PATENT ASSIGNEE(S): Cellfree Sciences Co., Ltd., Japan  
 SOURCE: PCT Int. Appl., 35 pp.  
 CODEN: PIXXD2  
 DOCUMENT TYPE: Patent  
 LANGUAGE: Japanese  
 FAMILY ACC. NUM. COUNT: 1  
 PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2006051901	A1	20060518	WO 2005-JP20715	20051111
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BW, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KM, KN, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, LY, MA, MD, MG, MK, MN, MW, MX, MZ, NA, NG, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, SM, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW RW: AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IS, IT, LI, LU, LV, MC, NL, PL, PT, RO, SE, SI, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG, BW, GH, GM, KE, LS, MW, MZ, NA, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM JP 2008029203 A 20080214 JP 2004-329794 20041112 PRIORITY APPLN. INFO.: JP 2004-329794 A 20041112				

ED Entered STN: 21 May 2006

AB The cell-free protein synthesis comprises using directly the transcription products without the purification of the mRNA. The method is highly efficient and easy. Preparation of S-30 fraction of wheat germ of the cell-free translation was shown.

REFERENCE COUNT: 6 THERE ARE 6 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 67 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN

ACCESSION NUMBER: 2006:469852 CAPLUS Full-text

DOCUMENT NUMBER: 144:447592

TITLE: Cell-free protein synthesizing bioreactor apparatus using

repetitive overlaying or repetitive batch-supplying system

INVENTOR(S): Endo, Yaeta; Sawasaki, Tatsuya; Ogasawara, Tomio

PATENT ASSIGNEE(S): Cellfree Sciences Co., Ltd., Japan

SOURCE: PCT Int. Appl., 44 pp.

CODEN: PIXXD2

DOCUMENT TYPE: Patent

LANGUAGE: Japanese

FAMILY ACC. NUM. COUNT: 1

PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2006051908	A1	20060518	WO 2005-JP20727	20051111
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BW, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KM, KN, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, LY, MA, MD, MG, MK, MN, MW, MX, MZ, NA, NG, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, SM, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW				
RW: AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IS, IT, LI, LU, LV, MC, NL, PL, PT, RO, SE, SI, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG, BW, GH, GM, KE, LS, MW, MZ, NA, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
JP 2008029204	A	20080214	JP 2004-329798	20041112
PRIORITY APPLN. INFO.:			JP 2004-329798	A 20041112

ED Entered STN: 19 May 2006

AB A novel cell free protein synthesis system that has been improved for rapid and simple operation by omitting the use of complicated compartments of membrane filtration or column chromatogs. is provided. The bioreactor uses repetitive overlaying or repetitive batch-supplying system for operation. The system can monitor the reaction rate in its reaction chamber and the reaction rate is controlled by the supply rate (volume/s) of new reaction batch in a continuous or an discontinuous manner. A new reaction batch is designed to be added (overlayed) to (on) the reaction mixture and mixed in the chamber upon the detection of the reduced synthesis rate (at 10 min .apprx. 10 h time intervals). The reaction batch contain purified or coarse post-transcriptional mRNA fraction and wheat germ lysate (endospem and inhibitor protein free and controlled sugar-phosphate levels). Condensation mechanism is used to remove reaction byproducts and excess reagents such as magnesium ion and nucleotides. Production of model protein (GFP) by the system (2 .apprx. 4 time repetitive reactions) was 1.1 .apprx. 1.8 mg/mL reaction mixture

REFERENCE COUNT: 14 THERE ARE 14 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 68 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN

ACCESSION NUMBER: 2006:240332 CAPLUS Full-text

DOCUMENT NUMBER: 145:119893

TITLE: Sequence-based analyses of biosynthesis rate limiting factors in wheat germ cell-free system  
 AUTHOR(S): Fujita, Naoya; Kinoshita, Kengo; Seki, Mutoaki; Sawasaka, Tatsuya; Nakai, Kenta; Shinozaki, Kazuo; Endo, Yaeta  
 CORPORATE SOURCE: Human Genome Center, Institute of Medical Science, University of Tokyo, 4-6-1 Shirokanedai, Minato-ku, Tokyo, 108-8639, Japan  
 SOURCE: Genome Informatics 2005, International Conference, Poster and Software Demonstrations, 16th, Yokohama, Japan, Dec. 19-21, 2005 (2005), P095/1-P095/2. Universal Academy Press: Tokyo, Japan.  
 CODEN: 69HXB6  
 DOCUMENT TYPE: Conference  
 LANGUAGE: English  
 ED Entered STN: 17 Mar 2006

AB The production of proteins is essential for their structural and functional analyses in the post-genome era. A wheat germ cell-free system is a useful method for this purpose. This system is able to produce proteins from various sources' mRNAs using the translational machinery from wheat germs. One improvement made in this system is the use of only one 5'-UTR, which is important in biosynthesis initiation, and one 3'-UTR for the expression of all coding sequences. However, depending on the coding sequences, the range of yields observed is very wide. The causes of the yield variation based on the protein sequences were investigated using a dataset consisting of 425 protein kinases from Arabidopsis thaliana. Based on the results obtained, two yield decreasing factors were identified: disorder and coiled coil.  
 REFERENCE COUNT: 3 THERE ARE 3 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 69 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN  
 ACCESSION NUMBER: 2006:117096 CAPLUS Full-text  
 DOCUMENT NUMBER: 144:186045  
 TITLE: Nucleic acid sequences having translation enhancement activity and use  
 INVENTOR(S): Sawasaka, Tatsuya; Endo, Yaeta; Kamura, Nami  
 PATENT ASSIGNEE(S): Cellfree Sciences Co., Ltd., Japan  
 SOURCE: U.S. Pat. Appl. Publ., 16 pp.  
 CODEN: USXXCO  
 DOCUMENT TYPE: Patent  
 LANGUAGE: English  
 FAMILY ACC. NUM. COUNT: 1  
 PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
US 20060029999	A1	20060209	US 2005-53594	20050208
JP 2006042676	A	20060216	JP 2004-227866	20040804
PRIORITY APPLN. INFO.:			JP 2004-227866	A 20040804

ED Entered STN: 09 Feb 2006  
 AB The present invention provides a polynucleotide comprising a nucleic acid sequence having an activity of regulating the translation efficiency of a template in a cell-free protein synthesis system and also provides a method for utilizing the same, etc. Protein synthesis is carried out by a translation template containing a polynucleotide comprising a nucleic acid sequence which is to be an object to be selected, a polyribosome fraction is prepared from the reaction solution and a nucleic acid sequence bonding to a ribosome is analyzed whereupon a selection is done.

L7 ANSWER 70 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN  
 ACCESSION NUMBER: 2005:1013930 CAPLUS Full-text  
 DOCUMENT NUMBER: 144:306950

TITLE: The wheat germ cell-free expression system: Methods for high-throughput materialization of genetic information  
 AUTHOR(S): Sawasaki, Tatsuya; Gouda, Mudeppa D.; Kawasaki, Takayasu; Tsuboi, Takafumi; Tozawa, Yuzuru; Takai, Kazuyuki; Endo, Yaeta  
 CORPORATE SOURCE: Cell-Free Science and Technology Research Center, Ehime University, Matsuyama, Japan  
 SOURCE: Methods in Molecular Biology (Totowa, NJ, United States) (2005), 310 (Chemical Genomics), 131-144  
 CODEN: MMBIED; ISSN: 1064-3745  
 PUBLISHER: Humana Press Inc.  
 DOCUMENT TYPE: Journal  
 LANGUAGE: English  
 ED Entered STN: 20 Sep 2005  
 AB This chapter contains protocols for high-throughput protein production based on the cell-free system prepared from eukaryote wheat embryos.  
 REFERENCE COUNT: 18 THERE ARE 18 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 71 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN  
 ACCESSION NUMBER: 2005:612456 CAPLUS Full-text  
 DOCUMENT NUMBER: 143:128797  
 TITLE: Regulation of ATP mediated phosphorylation of carbohydrate for increase the efficiency of cell-free protein synthesis  
 INVENTOR(S): Endo, Yaeta; Ogasawara, Tomio  
 PATENT ASSIGNEE(S): Cellfree Sciences Co., Ltd., Japan  
 SOURCE: PCT Int. Appl., 39 pp.  
 CODEN: PIXXD2  
 DOCUMENT TYPE: Patent  
 LANGUAGE: Japanese  
 FAMILY ACC. NUM. COUNT: 1  
 PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2005063979	A1	20050714	WO 2004-JP18928	20041217
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BW, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NA, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW, RW: BW, GH, GM, KE, LS, MW, MZ, NA, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IS, IT, LT, LU, MC, NL, PL, PT, RO, SE, SI, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG				
US 20070141661	A1	20070621	US 2006-596538	20060710
PRIORITY APPLN. INFO.:			JP 2003-434080	A 20031226
			WO 2004-JP18928	W 20041217

ED Entered STN: 15 Jul 2005  
 AB This invention provides a cell extract to increase the efficiency of cell-free protein synthesis by identifying and removing inhibitory and unstable contaminants in various existing cell exts.. The ATP-mediated carbohydrate phosphorylation system in the cell extract is regulated. The process consists of 1 removal of monosaccharides; 2 removal of phosphorylated saccharides; 3 regulation of the formation of monosaccharides from polysaccharides; and 4 regulation of the formation of phosphorylated saccharides from monosaccharides.  
 REFERENCE COUNT: 4 THERE ARE 4 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 72 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN

ACCESSION NUMBER: 2005:478393 CAPLUS Full-text  
 DOCUMENT NUMBER: 144:145607  
 TITLE: in vitro translation systems  
 AUTHOR(S): Sawasaki, Tatsuya; Endo, Yaeta  
 CORPORATE SOURCE: Research Center of Cell-Free Bio-Science and Bio-Engineering, Ehime University, Japan  
 SOURCE: Shokubutsu Saibo Kogaku Shirizu (2005), 21(Moderu Shokubutsu no Jikken Purotokoru (3rd Edition)), 230-234  
 CODEN: SSKSFR  
 PUBLISHER: Shujunsha  
 DOCUMENT TYPE: Journal; General Review  
 LANGUAGE: Japanese  
 ED Entered STN: 06 Jun 2005

AB A review. A typical protocol of the recombinant protein production in the cell free protein translation system using wheat germ extract was presented. The coverage of the procedures included the preparation of the wheat germ extract, the preparation of the template plasmid DNA (pEU: plasmid of Ehime University) for expressing recombinant fusion protein with GST, mRNA purification, in vitro protein synthesis and the purification by using an affinity resin.

L7 ANSWER 73 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN

ACCESSION NUMBER: 2005:300592 CAPLUS Full-text  
 DOCUMENT NUMBER: 142:353880  
 TITLE: Producing antigens in wheat germ cell-free protein synthesis system for vaccine preparation  
 INVENTOR(S): Endo, Yaeta; Tsuboi, Takafumi; Torii, Motomi; Sawasaki, Tatsuya  
 PATENT ASSIGNEE(S): Cellfree Sciences Co., Ltd., Japan  
 SOURCE: PCT Int. Appl., 39 pp.  
 CODEN: PIXXD2  
 DOCUMENT TYPE: Patent  
 LANGUAGE: Japanese  
 FAMILY ACC. NUM. COUNT: 1  
 PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2005030954	A1	20050407	WO 2004-JP13918	20040924
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BW, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NA, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW RW: BW, GH, GM, KE, LS, MW, MZ, NA, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PL, PT, RO, SE, SI, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG				
EP 1669448	A1	20060614	EP 2004-788077	20040924
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, FI, RO, CY, TR, BG, CZ, EE, HU, PL, SK US 20060233789 A1 20061019 US 2006-572139 20060411 JP 2003-333659 A 20030925 WO 2004-JP13918 W 20040924				
PRIORITY APPLN. INFO.: ED Entered STN: 07 Apr 2005				

AB Provided here is a means of producing antigens retaining its native antigenicity by using a cell-free protein synthesis means. In particular, it provides a means of producing antigens without being ruled by codon usage via, for example, from an AT-rich gene. Antibodies produced with the vaccine, and use as diagnostic agent, and kit are also disclosed. Screening of

antimalarials is also disclosed. Antigens retaining its antigenicity (in particular, a malaria antigens useful in producing malaria vaccines) were successfully prep'd. with wheat germ cell-free protein synthesis system. Transmission-blocking vaccines target the sexual stages of the malaria parasite and prevent further development within the mosquito vector halting the transmission of the parasite. Zygote/ookinetes are potential targets of antibodies inhibiting oocyst development in the mosquito midgut and rendering mosquitoes non-infectious. DNA vaccine constructs were developed expressing Pvs25 and Pvs28 (Plasmodium vivax zygote/ookinete surface proteins). Antibodies produced in mice after immunization recognized resp. antigens, and these antibodies when tested in assay were potent blockers of P. vivax transmission.

REFERENCE COUNT: 8 THERE ARE 8 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 74 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN  
 ACCESSION NUMBER: 2005:297489 CAPLUS Full-text  
 DOCUMENT NUMBER: 142:332432  
 TITLE: Preparation of cell extracts for cell-free translation  
 INVENTOR(S): Tozawa, Yuzuru; Kanno, Takuya; Endo, Yaeta; Dohi, Naoki;  
 Koga, Hirohisa  
 PATENT ASSIGNEE(S): Zoegene Corporation, Japan  
 SOURCE: Jpn. Kokai Tokkyo Koho, 14 pp.  
 CODEN: JKXXAF  
 DOCUMENT TYPE: Patent  
 LANGUAGE: Japanese  
 FAMILY ACC. NUM. COUNT: 1  
 PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
JP 2005087208	A	20050407	JP 2004-228349	20040804
PRIORITY APPLN. INFO.:			JP 2003-289874	A 20030808

ED Entered STN: 07 Apr 2005

AB Cells are segmented, and mixed with extraction solvents to prepare cell exts. In the extraction process, an activation agents selected from aluminum oxide, activated aluminum oxide, silica gel, etc., is added for activation of the cell exts. The cells are preferably plant germ cells, especially wheat germ cells. The segmentation is achieved by impact or cutting. The method provides enhanced cell-free translation with high efficiency. Preparation of wheat germ extract by Waring blender, activation of the wheat germ extract with aluminum oxide, and enhanced cell-free translation of green fluorescence protein (GFP) were shown.

L7 ANSWER 75 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN  
 ACCESSION NUMBER: 2005:239233 CAPLUS Full-text  
 DOCUMENT NUMBER: 142:291317  
 TITLE: Novel high throughput screening method of drug for physiologically active protein  
 INVENTOR(S): Endo, Yaeta; Sawasaki, Tatsuya  
 PATENT ASSIGNEE(S): Cellfree Sciences Co., Ltd., Japan  
 SOURCE: PCT Int. Appl., 47 pp.  
 CODEN: PIXXD2  
 DOCUMENT TYPE: Patent  
 LANGUAGE: Japanese  
 FAMILY ACC. NUM. COUNT: 1  
 PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
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WO 2005024428 A1 20050317 WO 2004-JP13071 20040908  
 W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BW, BY, BZ, CA, CH, CN,  
 CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID,  
 IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN,  
 MW, MX, MZ, NA, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RU, SC, SD, SE,  
 SG, SK, SL, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW  
 RW: BW, GH, GM, KE, LS, MW, MZ, NA, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ,  
 BY, KG, KZ, MD, RU, TJ, TM, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR,  
 HU, IE, IT, LU, MC, NL, PL, PT, RO, SE, SI, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN,  
 GQ, GW, ML, MR, NE, SN, TD, TG  
 EP 1669759 A1 20060614 EP 2004-787754 20040908  
 R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE,  
 SI, FI, RO, CY, TR, BG, CZ, EE, HU, PL, SK  
 US 20060177813 A1 20060810 US 2006-571081 20060331  
 JP 2003-316081 A 20030908  
 PRIORITY APPLN. INFO.: WO 2004-JP13071 W 20040908

ED Entered STN: 18 Mar 2005

AB A safe and rapid means for screening a drug (in particular, an inhibitor) for  
 a physiol. active protein is provided, with which a system for synthesizing a  
 physiol. active protein sustaining its activity is constructed by utilizing a  
 wheat germ extract cell-free protein synthesis system among cell-free protein  
 synthesis means. As a typical example of using this synthesis system, a  
 system for screening a SARS 3CLpro inhibitor is constructed.

REFERENCE COUNT: 7 THERE ARE 7 CITED REFERENCES AVAILABLE FOR THIS  
 RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 76 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN

ACCESSION NUMBER: 2004:965399 CAPLUS Full-text

DOCUMENT NUMBER: 141:391506

TITLE: Automatic process and apparatus for high throughput cell-  
 free protein synthesis utilizing membrane filter concentration

INVENTOR(S): Endo, Yae; Sawasaki, Tatsuya; Ogasawara, Tomio;

Morishita, Riyo; Saeki, Mihoro; Sato, Tomohisa; Kitamoto, Aya

PATENT ASSIGNEE(S): Japan

SOURCE: PCT Int. Appl., 81 pp.

CODEN: PIXXD2

DOCUMENT TYPE: Patent

LANGUAGE: Japanese

FAMILY ACC. NUM. COUNT: 1

PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2004097014	A1	20041111	WO 2004-JP5912	20040423
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BW, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NA, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW RW: BW, GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PL, PT, RO, SE, SI, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG				
JP 2006042601	A	20060216	JP 2003-281500	20030729
AU 2004234669	A1	20041111	AU 2004-234669	20040423
CA 2522927	A1	20041111	CA 2004-2522927	20040423
EP 1619247	A1	20060125	EP 2004-729258	20040423
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR, BG, CZ, EE, HU, PL, SK, HR				



CN 1777674	A	20060524	CN 2004-80010994	20040423
US 20060257997	A1	20061116	US 2005-554434	20051214
PRIORITY APPLN. INFO.:			JP 2003-122930	A 20030425
			JP 2003-281500	A 20030729
			WO 2004-JP5912	W 20040423

ED Entered SIN: 12 Nov 2004

AB A method and automated apparatus for high throughput in vitro synthesis of biopolymers such as proteins or RNA, are disclosed. The cell-free synthesis system comprises the following means: (1) a template material, a substrate and a reaction solution are contacted together and introduced into a synthesis reaction system. (2) before or after almost lowering the synthesis speed, or almost ceasing the synthesis reaction, or during the same, the reaction system is taken out from the synthesis reaction system and the solution is diluted (3) following the dilution, it is concd. (4) the reaction system is returned into the synthesis reaction system. An alternative means is as follows. (1) a template material, a substrate and a reaction solution are contacted together and introduced into a synthesis reaction system. (2) before or after almost lowering the synthesis speed, or almost ceasing the synthesis reaction, or during the same, the reaction system is taken out from the synthesis reaction system and the solution is concentrated (3) following the concn., it is diluted (4) the diluted reaction system is returned into the synthesis reaction system. Concentration is accomplished with a membrane filter, centrifuge, and/or a suction pump. Information processing program for the automated apparatus is claimed.

REFERENCE COUNT: 10 THERE ARE 10 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 77 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN

ACCESSION NUMBER: 2004:678409 CAPLUS Full-text  
DOCUMENT NUMBER: 141:170409  
TITLE: Automatic process for protein synthesis and apparatus therefor  
INVENTOR(S): Endo, Yae; Sasaki, Tatsuya  
PATENT ASSIGNEE(S): Japan  
SOURCE: PCT Int. Appl., 41 pp.  
CODEN: PIXXD2  
DOCUMENT TYPE: Patent  
LANGUAGE: Japanese  
FAMILY ACC. NUM. COUNT: 1  
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
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WO 2004070047	A1	20040819	WO 2004-JP1364	20040210
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BW, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NA, NI				
RW: BW, GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PT, RO, SE, SI, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG				

PRIORITY APPLN. INFO.:

ED Entered SIN: 19 Aug 2004

AB In cell-free protein synthesis, the transcription template is precipitated and dried while the supernatant is removed. The transcription template is easily solubilized in the cell-free protein synthesis system. The low-mol. weight protein synthesis inhibitors in the plant seed germ extract such as wheat germ extract and albumen is removed prior to the cell-free protein synthesis. The method is highly efficient; the yield of mRNA is high; and loss of mRNA is minimal. An automated cell-free protein synthesis apparatus

comprising temperature control system, sample supply system, precipitation means, supernatant removal system, drying system. etc., is also given.

L7 ANSWER 78 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN

ACCESSION NUMBER: 2004:80730 CAPLUS Full-text  
DOCUMENT NUMBER: 140:144709  
TITLE: Single chain antibody comprising light and heavy chains coupled by a labeled linker for antigen immunoassay chip and test kit  
INVENTOR(S): Endo, Naeta; Kawasaki, Takayasu; Sawasaki, Tatsuya  
PATENT ASSIGNEE(S): Japan  
SOURCE: PCT Int. Appl., 63 pp.  
CODEN: PIXXD2  
DOCUMENT TYPE: Patent  
LANGUAGE: Japanese  
FAMILY ACC. NUM. COUNT: 1  
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2004009639	A1	20040129	WO 2003-JP9140	20030718
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MX, MZ, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, SY, TJ, TM, TN, TR, TT, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW				
R: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, YU, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PT, RO, SE, SI, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG				
CA 2492996	A1	20040129	CA 2003-2492996	20030718
AU 2003248081	A1	20040209	AU 2003-248081	20030718
EP 1541588	A1	20050615	EP 2003-765324	20030718
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR, BG, CZ, EE, HU, SK				
US 20060172344	A1	20060803	US 2005-522000	20050223
PRIORITY APPLN. INFO.:			JP 2002-210067	A 20020718
			WO 2003-JP9140	W 20030718

ED Entered STN: 01 Feb 2004

AB It is intended to provide a single chain antibody sustaining an activity of specifically binding to an antigen and a labeled single chain antibody composed of the single chain antibody and a label bonded thereto. More specifically, the labeled single chain antibody as described above can be produced by bonding a label to the linker moiety of the single chain antibody. This antibody is produced by using a wheat germ-origin cell free protein synthesis system in a less reductive state where a disulfide bond in a mol. can be maintained. Further, the antibody is bonded to a solid phase via the label to thereby produce an immobilized single chain antibody. Also, an antigen-antibody reaction is analyzed with the use of this immobilized single chain antibody.

REFERENCE COUNT: 13 THERE ARE 13 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 79 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN

ACCESSION NUMBER: 2003:938880 CAPLUS Full-text  
DOCUMENT NUMBER: 139:394969  
TITLE: Selection of germs from plant seeds, manufacture of the germ extracts, and method, solutions, and kits for cell-free protein synthesis using the extracts  
INVENTOR(S): Dohi, Naoki; Morisada, Shigeo; Iwahashi, Shigeo; Endo,

Yasta

PATENT ASSIGNEE(S): Japan  
SOURCE: Jpn. Kokai Tokkyo Koho, 12 pp.  
CODEN: JKXXAF  
DOCUMENT TYPE: Patent  
LANGUAGE: Japanese  
FAMILY ACC. NUM. COUNT: 1  
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
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JP 2003339395	A	20031202	JP 2002-152245	20020527
PRIORITY APPLN. INFO.:			JP 2002-152245	20020527

ED Entered STN: 02 Dec 2003

AB Intact germs, from which exts. are prepared for cell-free protein synthesis, are selected by milling plant seeds using an impact mill at impact velocity 25-70 m/s and separating germs from the milled product. Germ exts. are manufactured by pulverizing the above selected germs and extracting the germs. Also claimed are a cell-free protein synthesis method using the germ exts., solns. containing the germ exts., ATP, GTP, creatine phosphate, creatine kinase, L-amino acids, K+, and Mg2+, and reagent kits containing the germ exts. or the solns. Wheat was hammer-milled at impact velocity 35 m/s and sieved to recover a 0.7-1.1-mm germ-containing fraction. The fraction was fed to a pneumatic classifier to remove bran, endoderm, etc. Germs having germinating capacity were separated from the fraction by heavy medium separation using CCl4/cyclohexane (2.4:1), dried, and remaining brans were removed by winnowing. The crude germ fraction was further separated into germs and seed coat/endosperm by a method based on difference in color. The separated germs were ultrasonically cleaned in H2O at 4° and further ultrasonically cleaned in Nonidet solution to completely remove endosperm component. The cleaned germs were extracted with a soln. containing HEPES-KOH, AcOK, Mg(OAc)2, CaCl2, L-amino acids, and dithiothreitol in a Waring blender. The mixture was centrifuged and the supernatant was gel-filtered, and the filtrate was centrifuged again to give wheat germ extract. Synthesis of green fluorescent protein using the extract was also shown.

L7 ANSWER 80 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN

ACCESSION NUMBER: 2003:936309 CAPLUS Full-text  
DOCUMENT NUMBER: 139:394968  
TITLE: Manufacture of germ extracts, solution containing them,  
and method and kit for cell-free protein synthesis using the solution  
INVENTOR(S): Dohi, Naoki; Morisada, Shigeo; Iwahashi, Shigeo; Endo,  
Yasta

PATENT ASSIGNEE(S): Japan  
SOURCE: Jpn. Kokai Tokkyo Koho, 12 pp.  
CODEN: JKXXAF  
DOCUMENT TYPE: Patent  
LANGUAGE: Japanese  
FAMILY ACC. NUM. COUNT: 1  
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
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JP 2003339394	A	20031202	JP 2002-152236	20020527
PRIORITY APPLN. INFO.:			JP 2002-152236	20020527

ED Entered STN: 02 Dec 2003

AB In manufacture of germ exts. by milling plant seeds, separating germs, washing the germs, grinding the germs, and extracting the germs, the germs are contacted with nonchlorine solvents prior to grinding to remove lipids. Also

claimed are cell-free protein synthesis solns. containing the germ exts., method to cell-free protein synthesis using the solns., and cell-free protein synthesis kits containing the solns. Wheat germs were recovered from milled wheat by sieving and winnowing and fed to an electromagnetic feeder having an vibrator to sep. crude germ fraction. The crude fraction was further separated into germs and seed coat/endosperm by a method based on difference in color. The separated germs were treated with EtOH for 5 min, dried, ultrasonically cleaned in H<sub>2</sub>O at 4°, and further ultrasonically cleaned in Nonidet solution. The cleaned germs were extracted with a solution containing HEPES-KOH, AcOK, Mg(OAc)<sub>2</sub>, CaCl<sub>2</sub>, L-amino acids, and dithiothreitol in a Waring blender. The mixture was centrifuged and the supernatant was gel-filtered, and the filtrate was centrifuged again to give wheat germ extract. Synthesis of green fluorescent protein using the extract was also shown.

L7 ANSWER 81 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN

ACCESSION NUMBER: 2003:913316 CAPLUS Full-text  
DOCUMENT NUMBER: 139:360695  
TITLE: Lyophilized preparation for cell-free protein synthesis  
INVENTOR(S): Endo, Yaeta; Ogasawara, Tomio  
PATENT ASSIGNEE(S): Japan  
SOURCE: PCT Int. Appl., 32 pp.  
CODEN: PIXXD2  
DOCUMENT TYPE: Patent  
LANGUAGE: Japanese  
FAMILY ACC. NUM. COUNT: 1  
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2003095661	A1	20031120	WO 2003-JP5656	20030506
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, GR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NI, NO, NZ, OM, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW				
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PT, RO, SE, SI, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG				
AU 2003235844	A1	20031111	AU 2003-235844	20030506
CA 2485827	A1	20031120	CA 2003-2485827	20030506
EP 1550728	A1	20050706	EP 2003-721030	20030506
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR, BG, CZ, EE, HU, SK				
US 20050153390	A1	20050714	US 2004-514855	20041221
PRIORITY APPLN. INFO.:			JP 2002-138828	A 20020514
			WO 2003-JP5656	W 20030506

ED Entered STN: 21 Nov 2003

AB The lyophilized cell-free protein synthesis system has comparable activity to that prepared by low-temperature preservation. The cell-free protein synthesis system contains lower deliquescent substances such as potassium acetate. The deliquescent substances amount to ≤0.01 weight% based on 1 weight% protein in the cell-free protein synthesis system. The low-mol. weight protein synthesis inhibitors are removed in the presence of high-energy phosphate compds. such as ATP.

REFERENCE COUNT: 9 THERE ARE 9 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 82 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN

ACCESSION NUMBER: 2003:832735 CAPLUS Full-text  
 DOCUMENT NUMBER: 141:19197  
 TITLE: High-throughput functional and structural analysis of  
 proteins based on cell-free protein synthetic technology  
 AUTHOR(S): Endo, Yaeta; Sawasaki, Tatsuya  
 CORPORATE SOURCE: Faculty of Engineering, Department of Applied Chemistry,  
 Ehime University, Matsuyama, 790-8577, Japan  
 SOURCE: Biobench (2003), 3(5), 22-24  
 CODEN: BIOBC8; ISSN: 1346-5376  
 PUBLISHER: Yodosha  
 DOCUMENT TYPE: Journal; General Review  
 LANGUAGE: Japanese  
 ED Entered STN: 24 Oct 2003

AB A review. Advantages of the use of cell-free protein translation system to  
 prepare stable protein samples efficiently for structural analyses were  
 discussed. Tech. break through to improve production efficiency of wheat germ  
 lysate cell-free system was described. Application of the technol. to high-  
 throughput screening for functional and structural anal. of proteins was  
 discussed. Development of automatized protein synthesis robot system was also  
 discussed.

L7 ANSWER 83 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN

ACCESSION NUMBER: 2003:697078 CAPLUS Full-text  
 DOCUMENT NUMBER: 139:210387  
 TITLE: Reaction solution for and preparation of cell-free protein  
 synthesis system for manufacture of protein and antibody  
 INVENTOR(S): Endo, Yaeta; Kawasaki, Takayasu; Sawasaki, Tatsuya  
 PATENT ASSIGNEE(S): Japan  
 SOURCE: PCT Int. Appl., 62 pp.  
 CODEN: PIXXD2  
 DOCUMENT TYPE: Patent  
 LANGUAGE: Japanese  
 FAMILY ACC. NUM. COUNT: 1  
 PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2003072796	A1	20030904	WO 2003-JP2313	20030228
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PT, SE, SI, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG CA 2477440 A1 20030904 CA 2003-2477440 20030228 AU 2003211458 A1 20030909 AU 2003-211458 20030228 EP 1489188 A1 20041222 EP 2003-743063 20030228 R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR, BG, CZ, EE, HU, SK US 20050148046 A1 20050707 US 2004-506127 20041104 US 7273615 B2 20070925 PRIORITY APPLN. INFO.: JP 2002-53161 A 20020228 WO 2003-JP2313 W 20030228				

ED Entered STN: 05 Sep 2003

AB The cell-free protein synthesis system (redox potential, -100 to 0 mV) is  
 useful for formation of intramol. disulfide linkage and folding of proteins,

especially antibodies. The cell-free protein synthesis system contains  $\geq 1$  reductants selected from dithiothreitol, 2-mercaptoethanol, and glutathione. It further contains catalysts such as disulfide isomerase for disulfide exchange and formation of proper disulfide linkage. Manufacture of single-stranded antibody to Salmonella O-antigen with a wheat germ cell-free protein synthesis system was shown.

REFERENCE COUNT: 15 THERE ARE 15 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 84 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN

ACCESSION NUMBER: 2003:610649 CAPLUS Full-text  
DOCUMENT NUMBER: 139:145563  
TITLE: "Ready-made" wheat germ cell-free protein synthesis system  
INVENTOR(S): Endo, Yaeta  
PATENT ASSIGNEE(S): Japan  
SOURCE: PCT Int. Appl., 55 pp.  
CODEN: PIXXD2  
DOCUMENT TYPE: Patent  
LANGUAGE: Japanese  
FAMILY ACC. NUM. COUNT: 1  
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2003064672	A1	20030807	WO 2003-JP975	20030131
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW				
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PT, SE, SI, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG				
CA 2483332	A1	20030807	CA 2003-2483332	20030131
EP 1479776	A1	20041124	EP 2003-734897	20030131
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR, BG, CZ, EE, HU, SK				
JP 3675804	B2	20050727	JP 2003-564263	20030131
US 20050064592	A1	20050324	US 2004-503259	20041115
PRIORITY APPLN. INFO.:			JP 2002-23141	A 20020131
			WO 2003-JP975	W 20030131

ED Entered STN: 08 Aug 2003

AB The low-mol.-weight inhibitors (mol.-weight,  $\leq 14000$  dalton) to protein synthesis in the cell-free protein synthesis germ extract, especially wheat germ extract, is removed by dialysis using regenerated cellulose membrane or by gel filtration. The dialysis was done in the presence of high-energy phosphates such as ATP and/or amino acids for stabilization of the cell-free protein synthesis system. Removal of the low-mol.-weight inhibitors from wheat germ extract cell-free protein synthesis system with Spectrapor 6 (Spectrum Medical Ind., Inc.) was shown.

REFERENCE COUNT: 18 THERE ARE 18 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 85 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN

ACCESSION NUMBER: 2003:610648 CAPLUS Full-text  
DOCUMENT NUMBER: 139:145562  
TITLE: Germ extract for cell-free protein synthesis and process for producing the same  
INVENTOR(S): Endo, Yaeta; Dohi, Naoki; Nakagawa, Makoto

PATENT ASSIGNEE(S): Japan  
 SOURCE: PCT Int. Appl., 45 pp.  
 CODEN: PIXXD2  
 DOCUMENT TYPE: Patent  
 LANGUAGE: Japanese  
 FAMILY ACC. NUM. COUNT: 1  
 PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2003064671	A1	20030807	WO 2003-JP995	20030131
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PT, SE, SI, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG				
CA 2474466	A1	20030807	CA 2003-2474466	20030131
EP 1477566	A1	20041117	EP 2003-734900	20030131
EP 1477566	B1	20070926		
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR, BG, CZ, EE, HU, SK				
JP 3746780	B2	20060215	JP 2003-564262	20030131
AT 374258	T	20071015	AT 2003-734900	20030131
US 20050042305	A1	20050224	US 2004-503271	20041025
JP 2005211074	A	20050811	JP 2005-62941	20050307
JP 3956314	B2	20070808		
PRIORITY APPLN. INFO.:				
			JP 2002-23138	A 20020131
			JP 2002-23139	A 20020131
			JP 2002-23140	A 20020131
			JP 2003-564262	A3 20030131
			WO 2003-JP995	W 20030131

ED Entered STN: 08 Aug 2003

AB Germ exts. of wheat, barley, rice, and corn are prepared by impacting, and/or cutting in the presence/absence of extraction solvent for stable and high-efficiency cell-free protein synthesis system. The germ exts. have reduced RNase activity ( $\leq 10$  pg/ $\mu$ l). The DNA and total fatty acids in the germ exts. are 230  $\mu$ g/mL and  $\leq 0.03$  g/100g, resp., when the (optical d., O. D.) A260 = 90. It does not require the addition of t-RNA, and is low in impurities such as organelles, cell wall, etc. Manufacture of dihydrofolic acid dehydrogenase using the wheat germ cell-free protein synthesis system was shown.

REFERENCE COUNT: 23 THERE ARE 23 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 86 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN

ACCESSION NUMBER: 2003:532764 CAPLUS Full-text

DOCUMENT NUMBER: 139:80165

TITLE: Isolating nucleotide sequence regulating the translation efficiency in cell-free protein synthesis system

INVENTOR(S): Endo, Yaeta; Sawasaki, Tatsuya

PATENT ASSIGNEE(S): Japan

SOURCE: PCT Int. Appl., 89 pp.

CODEN: PIXXD2

DOCUMENT TYPE: Patent

LANGUAGE: Japanese

FAMILY ACC. NUM. COUNT: 1

PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2003056009	A1	20030710	WO 2002-JP13756	20021227
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW				
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, SI, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG				
CA 2471667	A1	20030710	CA 2002-2471667	20021227
AU 2002367144	A1	20030715	AU 2002-367144	20021227
EP 1466972	A1	20041013	EP 2002-790919	20021227
EP 1466972	B1	20070228		
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR, BG, CZ, EE, SK				
JP 3701292	B2	20050928	JP 2003-556526	20021227
AT 355366	T	20060315	AT 2002-790919	20021227
US 20040248140	A1	20041209	US 2004-500346	20040729
PRIORITY APPLN. INFO.:			JP 2001-396941	A 20011227
			WO 2002-JP13756	W 20021227

ED Entered STN: 11 Jul 2003

AB A method of preparing a polynucleotide containing a nucleotide sequence which regulate the translation efficiency of a template in a protein synthesis system, comprising (a) applying a template containing one or more arbitrary nucleotide sequences to a protein synthesis reaction system, (b) after reacting, recovering a polyribosome fraction from the liquid reaction mixture, and (c) collecting a polynucleotide contg. the nucleotide sequence in the template from the polyribosome fraction; novel polynucleotides regulating the translation efficiency obtained by the above method; a method of synthesizing a protein with the use of a template containing such a polynucleotide; and so on. Use of d. gradient centrifugation for collecting polyribosome fraction is claimed. Isolation of 27 57nt randomized sequences and 96 22nt randomized sequences are reported.

REFERENCE COUNT: 8 THERE ARE 8 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 87 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN

ACCESSION NUMBER: 2003:235544 CAPLUS [Full-text](#)

DOCUMENT NUMBER: 138:316222

TITLE: Cell-free protein synthesis by wheat germ extracts

AUTHOR(S): Tozawa, Yuzuru; Sawasaka, Tatsuya; Endo, Yaeta

CORPORATE SOURCE: Mitsubishi Kagaku Inst. Life Sci., Yokohama, 227-8502, Japan

SOURCE: Nippon Kessho Gakkaishi (2003), 45(1), 3-8

CODEN: NKEGAF; ISSN: 0369-4585

PUBLISHER: Nippon Kessho Gakkai

DOCUMENT TYPE: Journal; General Review

LANGUAGE: Japanese

ED Entered STN: 27 Mar 2003

AB A review. With the sequencing of the genomes of various species, attention has turned to the structure, properties, and functional activities of proteins. However, rapid progress in the area of proteomics is premised on the availability of sufficient amts. of a large number of proteins. Here we described a novel cell-free system from wheat embryos for the high-throughput screening/synthesis of gene products. Our system should open up many possibilities in the post-genome era.



L7 ANSWER 88 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN  
 ACCESSION NUMBER: 2003:123069 CAPLUS Full-text  
 DOCUMENT NUMBER: 138:148662  
 TITLE: Stabilization of nucleic acid template in cell-free  
 protein synthesis  
 INVENTOR(S): Sawasaki, Tatsuya; Ogasawara, Tomio; Endo, Yaeta  
 PATENT ASSIGNEE(S): Japan  
 SOURCE: Jpn. Kokai Tokkyo Koho, 18 pp.  
 CODEN: JKXXAF  
 DOCUMENT TYPE: Patent  
 LANGUAGE: Japanese  
 FAMILY ACC. NUM. COUNT: 1  
 PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
JP 2003047496	A	20030218	JP 2001-238697	20010807
PRIORITY APPLN. INFO.:			JP 2001-238697	20010807

ED Entered STN: 18 Feb 2003

AB Substrates for nucleic acid-degrading enzyme(s) are used in the cell-free protein synthesis system to stabilize the nucleic acid template. The substrates are selected from nucleic acids that does not have the starting codon AUG, that do not code for the protein of interest, and that have low transcription/translation activity in the cell-free protein synthesis system. Enhanced expression of GFP gene in a wheat germ cell-free protein synthesis system in the presence of AUG-free GFP RNA was shown.

L7 ANSWER 89 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN  
 ACCESSION NUMBER: 2002:906658 CAPLUS Full-text  
 DOCUMENT NUMBER: 137:366016  
 TITLE: Method for screening plant embryo for cell-free protein  
 synthesis, and method for producing embryo extract for cell-free protein synthesis  
 INVENTOR(S): Endo, Yaeta; Iwahashi, Shigeo; Nomura, Kazuo  
 PATENT ASSIGNEE(S): Japan  
 SOURCE: PCT Int. Appl., 37 pp.  
 CODEN: PIXXD2  
 DOCUMENT TYPE: Patent  
 LANGUAGE: Japanese  
 FAMILY ACC. NUM. COUNT: 3  
 PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2002095377	A1	20021128	WO 2002-JP4756	20020516
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZM, ZW				
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG				
JP 2002336797	A	20021126	JP 2001-149274	20010518
JP 2002338597	A	20021127	JP 2001-149275	20010518
AU 2002258191	A1	20021203	AU 2002-258191	20020516
JP 2003315328	A	20031106	JP 2002-141141	20020516
EP 1388733	A1	20040211	EP 2002-728068	20020516

R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE,  
SI, LT, LV, FI, RO, MK, CY, AL, TR

US 20040121462 A1 20040624 US 2004-478383 20040120  
JP 2001-149274 A 20010518  
JP 2001-149275 A 20010518  
JP 2002-43704 A 20020220  
WO 2002-JP4756 W 20020516

ED Entered STN: 29 Nov 2002

AB A method is provided for screening plant embryo for efficiently producing a germ extract for synthesizing a cell-free protein at a high synthesis efficiency. A method is also provided for producing a plant seed embryo extract for synthesizing a cell-free protein. Namely, the method for screening plant embryo for cell-free protein synthesis is characterized in that plant embryo is screened based on the optical information such as color information or image information, from a mixture containing embryo and ground albumen obtained by applying a mech. force to plant seeds (e.g., wheat, oat, rice). The method for producing a plant embryo extract is characterized in that the screened embryo is washed and finely ground, and afterwards, it is extracted

REFERENCE COUNT: 4 THERE ARE 4 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 90 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN

ACCESSION NUMBER: 2002:900830 CAPLUS Full-text

DOCUMENT NUMBER: 137:348861

TITLE: Method for preparing an embryo bud extract for cell-free protein synthesis, and cell-free protein synthesis method

INVENTOR(S): Endo, Yaeta; Ishibashi, Shigeo; Nomura, Kazuo

PATENT ASSIGNEE(S): Japan

SOURCE: Jpn. Kokai Tokkyo Koho, 9 pp.

CODEN: JKXXAF

DOCUMENT TYPE: Patent

LANGUAGE: Japanese

FAMILY ACC. NUM. COUNT: 3

PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
JP 2002338597	A	20021127	JP 2001-149275	20010518
WO 2002095377	A1	20021128	WO 2002-JP4756	20020516
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZM, ZW				
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG				
AU 2002258191	A1	20021203	AU 2002-258191	20020516
EP 1388733	A1	20040211	EP 2002-728068	20020516
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR				
CN 1511254	A	20040707	CN 2002-809706	20020516
US 20040121462	A1	20040624	US 2004-478383	20040120
PRIORITY APPLN. INFO.:				
			JP 2001-149274	A 20010518
			JP 2001-149275	A 20010518
			JP 2002-43704	A 20020220
			WO 2002-JP4756	W 20020516

ED Entered STN: 27 Nov 2002

AB An efficient method is provided for preparing an embryo bud extract for the cell-free protein synthesis with a high synthesis efficiency. A method is also provided for efficiently synthesizing a protein with the cell-free translation system on an industrial scale. The method for preparing an embryo bud extract comprises a process for grinding plant seeds (e.g., wheat, oat, rice, corn, spinach) and processes for selecting/washing/finely grinding/extracting the plant embryo bud. The selection of the plant embryo bud is characteristically performed using a color classifier.

L7 ANSWER 91 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN

ACCESSION NUMBER: 2002:416629 CAPLUS Full-text  
DOCUMENT NUMBER: 137:43626  
TITLE: Post-genome era. Development of cell-free protein synthesis system and its applications  
AUTHOR(S): Endo, Yaeta; Ogasawara, Tomio; Sawasaki, Tatsuya  
CORPORATE SOURCE: Department of Applied Chemistry, Faculty of Engineering, Ehime University, Matsuyama, 790-8577, Japan  
SOURCE: Kinoshi Kenkyu Kaishi (2002), Volume Date 2001, 40, 69-73  
CODEN: KIKKDD; ISSN: 0288-5867  
PUBLISHER: Kinoshi Kenkyukai  
DOCUMENT TYPE: Journal; General Review  
LANGUAGE: Japanese  
ED Entered STN: 04 Jun 2002

AB A review. Materialization of genetic information is an essential step toward modern biol. in both basic and applied fields today. We reported a novel system for massive production of gene products, which is based on wheat germ cell-free translation system. The methodol. consists of; (1) preparation of a highly efficient but also robust cell-free system, and (2) construction of a cell-free expression vector specialized for massive production of proteins. The method developed allowed to show that the system has high performance for materialization of genetic information directly from cDNA library. The possible applications of the system in the post-genome era are also discussed in this paper.

L7 ANSWER 92 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN

ACCESSION NUMBER: 2002:368678 CAPLUS Full-text  
DOCUMENT NUMBER: 136:336855  
TITLE: Process for producing germ extract  
INVENTOR(S): Endo, Yaeta; Yamamoto, Masaharu  
PATENT ASSIGNEE(S): Wakenyaku Co., Ltd., Japan  
SOURCE: PCT Int. Appl., 31 pp.  
CODEN: PIXXD2  
DOCUMENT TYPE: Patent  
LANGUAGE: Japanese  
FAMILY ACC. NUM. COUNT: 1  
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2002038790	A1	20020516	WO 2001-JP9778	20011108
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG				

JP 2002204689	A	20020723	JP 2000-341398	20001109
AU 2002012730	A5	20020521	AU 2002-12730	20011108
US 20040043088	A1	20040304	US 2003-416089	20030527
PRIORITY APPLN. INFO.:			JP 2000-340576	A 20001108
			JP 2000-341398	A 20001109
			WO 2001-JP9778	W 20011108

ED Entered STN: 18 May 2002

AB A efficient and low-cost process for producing a germ extract material characterized by involving the step of gently milling starting plant seeds to thereby eliminate the albumen of the seeds, the step of sieving the milled seeds under shaking to thereby recover fractions passing through 1.00-0.45 mm sieves, the step of eliminating the periderm of the seeds by winnowing, the step of recovering the suspension supernatant in water or an aqueous solution free from organic solvents (flotation), and the step of washing with water or an aqueous solution free from organic solvents; a germ extract material produced by this method; a germ extract obtained from this germ extract material; and a method of synthesizing a protein by using this germ extract. The method does not use organic solvent, prevents unnecessary protein synthesis associated with phys. damages of the seed, and is useful for com. manufacture of enzymes and antibodies. Synthesis of green fluorescence protein (GFP) with wheat germ extract was shown.

REFERENCE COUNT: 10 THERE ARE 10 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 93 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN

ACCESSION NUMBER: 2002:341321 CAPLUS Full-text

DOCUMENT NUMBER: 136:321725

TITLE: Use of apoprotein synthesized by cell-free system, and method for producing holoprotein by cell-free system

INVENTOR(S): Kuroita, Toshihiro; Kawakami, Fumikiyo; Kawamura, Yoshihisa; Nishikawa, Shigemichi; Endo, Yasta

PATENT ASSIGNEE(S): Toyobo Co., Ltd., Japan; Wakenyaku Co., Ltd.

SOURCE: Jpn. Kokai Tokkyo Koho, 12 pp.

CODEN: JKXXAF

DOCUMENT TYPE: Patent

LANGUAGE: Japanese

FAMILY ACC. NUM. COUNT: 1

PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
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JP 2002125698	A	20020508	JP 2000-320108	20001019
PRIORITY APPLN. INFO.:			JP 2000-320108	20001019

ED Entered STN: 08 May 2002

AB A method and a reagent are provided for conveniently and accurately detecting or quantitating a different kind of cofactor or coenzyme. A convenient and efficient method is also provided for producing a holoprotein by a cell-free protein synthesis system. The reagent for detecting or quantitating a cofactor or a coenzyme contains an apoprotein (e.g., apoenzyme) synthesized by a cell-free protein synthesis system. The method for producing a holoprotein (e.g., holoenzyme) is characterized by synthesizing the protein in the presence of a cofactor or a coenzyme by a cell-free protein synthesis system. An example is shown with the holoenzyme of sarcosine oxidase synthesized by the wheat germ cell-free extract in the presence of FAD.

L7 ANSWER 94 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN

ACCESSION NUMBER: 2002:341319 CAPLUS Full-text

DOCUMENT NUMBER: 136:336852

TITLE: Stabilization of freeze-dried cell extract for cell-free

protein synthesis

INVENTOR(S): Kuroita, Toshihiro; Kawakami, Fumikiyo; Kawamura, Yoshihisa; Nishikawa, Shigemichi; Endo, Yaeta  
 PATENT ASSIGNEE(S): Toyobo Co., Ltd., Japan; Wakenyaku Co., Ltd.  
 SOURCE: Jpn. Kokai Tokkyo Koho, 14 pp.  
 CODEN: JKXXAF

DOCUMENT TYPE: Patent  
 LANGUAGE: Japanese  
 FAMILY ACC. NUM. COUNT: 1  
 PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
JP 2002125693	A	20020508	JP 2000-320106	20001019
PRIORITY APPLN. INFO.:			JP 2000-320106	20001019

ED Entered STN: 08 May 2002

AB Inositol 0.3-3 weight% and polyalcs. 0.1-10, based on the weight of the protein in the cell ext., are useful for stabilization of freeze-dried cell extract for cell-free protein synthesis. The cell extract is obtained from wheat, barley, spinach, reticulocyte, Escherichia coli, etc. The cell extract may contain physiol. active substance selected from creatine kinase, pyruvate kinase, RNA polymerase, and chaperone proteins.

L7 ANSWER 95 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN

ACCESSION NUMBER: 2002:240998 CAPLUS Full-text  
 DOCUMENT NUMBER: 136:243573  
 TITLE: Methods for cell-free protein synthesis  
 INVENTOR(S): Endo, Yaeta; Sawasaki, Tatsuya; Ogasawara, Tomio  
 PATENT ASSIGNEE(S): Wakenyaku Co., Ltd., Japan  
 SOURCE: PCT Int. Appl., 33 pp.  
 CODEN: PIXXD2

DOCUMENT TYPE: Patent  
 LANGUAGE: Japanese  
 FAMILY ACC. NUM. COUNT: 1  
 PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2002024939	A1	20020328	WO 2001-JP7356	20010828
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW				
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG				
CA 2420984	A1	20020328	CA 2001-2420984	20010828
AU 2001080201	A	20020402	AU 2001-80201	20010828
EP 1316617	A1	20030604	EP 2001-958550	20010828
EP 1316617	B1	20070801		
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR				
CN 1449449	A	20031015	CN 2001-814793	20010828
JP 3768190	B2	20060419	JP 2002-529531	20010828
AU 2001280201	B2	20060713	AU 2001-280201	20010828
AT 368747	T	20070815	AT 2001-958550	20010828
CN 101092446	A	20071226	CN 2005-10090538	20010828
KR 749053	B1	20070813	KR 2003-702765	20030225

US 20030162246	A1	20030828	US 2003-344803	20030318
US 6869774	B2	20050322		
JP 2006136330	A	20060601	JP 2005-355513	20051209
PRIORITY APPLN. INFO.:			JP 2000-259186	A 20000829
			CN 2001-814793	A3 20010828
			JP 2002-529531	A3 20010828
			WO 2001-JP7356	W 20010828

ED Entered STN: 28 Mar 2002

AB A batch-type method for synthesizing a cell-free protein which is characterized by comprising, in a means of a reaction for synthesizing a cell-free protein, (1) bringing a synthetic reaction solution (a reaction phase) containing a biol. extract into contact directly with a substrate and an energy source supplying solution (a supplying phase) without separating by a semi-permeable membrane, thus continuously supplying the substrate and the energy source mols. of the supplying phase into the reaction phase by free diffusion via the contact interface and, at the same time, removal of the byproducts formed in the reaction phase into the supplying phase to thereby prolong the reaction time, and thus elevating the reaction efficiency; (2) pre-incubating (pre-heating) a reaction solution, containing a wheat germ extract, then diluting the synthesis reaction solution by adding a diluent solution to thereby prolong the synthetic reaction time, and thus elevating the synthesis efficiency; (3) re-supplying a substrate required in the synthesis of the protein (amino acid, ATP, GTP, creatine phosphate, etc.) and an energy source to the reaction mixture with the use of a gel filtration column or a semi-permeable membrane after ceasing the synthesis reaction and, at the same time, discontinuously taking off the byproducts formed by the reaction to thereby elevate the efficiency of the reaction. Manufacture of green fluorescence protein (GFP) with wheat germ cell-free protein synthesis system containing plasmid pEUL containing the gfp gene was shown. The production of the GFP was three time more than did the prior methods. The method is useful for automation of and robot development for post genomic study of gene function.

REFERENCE COUNT: 7 THERE ARE 7 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 96 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN

ACCESSION NUMBER: 2002:172088 CAPLUS Full-text  
DOCUMENT NUMBER: 136:227877  
TITLE: PCR primers for construction of transcription template for dilution batch-type cell-free protein synthesis system  
INVENTOR(S): Endo, Yasta; Sawasaki, Tatsuya; Ogasawara, Tomio  
PATENT ASSIGNEE(S): Wakenyaku Co., Ltd., Japan  
SOURCE: PCT Int. Appl., 66 pp.  
CODEN: PIXXD2  
DOCUMENT TYPE: Patent  
LANGUAGE: Japanese  
FAMILY ACC. NUM. COUNT: 1  
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
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WO 2002018586	A1	20020307	WO 2001-JP7357	20010828
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW				
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG				

AU 2001080202	A	20020313	AU 2001-80202	20010828
CA 2421012	A1	20030226	CA 2001-2421012	20010828
EP 1314781	A1	20030528	EP 2001-958551	20010828

R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR

AU 2001280202	B2	20061109	AU 2001-280202	20010828
US 20040121346	A1	20040624	US 2003-363372	20030227
			JP 2000-261638	A 20000830
			JP 2001-58404	A 20010302
			WO 2001-JP7357	W 20010828

PRIORITY APPLN. INFO.:

ED Entered STN: 08 Mar 2002

AB Primer sequences for construction of templates for a cell-free protein synthesis system using wheat germ extract are described. The 3'-terminal PCR primer contains sequences complementary to the sequence between the transcriptional termination sequence of a reporter gene (for example, a drug-resistance gene) of the vector and Ori. The 5'-terminal primer contains sequences complementary to part of the promoter sequence. Those two types of primers satisfy the requirement of not priming the transcription from a DNA constructed by using only of those primers alone. One of those having a sequence complementary to part of the RNA polymerase recognition site from the 5'-terminus of the promoter and another having a sequence complementary to part of the RNA polymerase recognition site from the 3'-terminus of the promoter are provided as primers for 5'-terminal PCR. GA or GAA sequences is ligated to those sequences, and further downstream, transcription initiation codon ATG, part of the target gene ORF. A histidine tag, glutathione-S-transferase (GST), or myb tag or epitope preparation sequence may also be used. The reaction mixture is to diluted achieve the optimal magnesium concentration. Cell-free protein synthesis using a modified mRNA having a tobacco mosaic virus (TMV)  $\Omega$  sequence ligated to the 5'-end and an untranslated region (UTR) ligated to the 3'-end, is described.

REFERENCE COUNT: 9 THERE ARE 9 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 97 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN

ACCESSION NUMBER: 2002:90248 CAPLUS [Full-text](#)  
DOCUMENT NUMBER: 136:114559  
TITLE: Labeling of protein in a cell- and embryo-free wheat germ protein synthesis system  
INVENTOR(S): Endo, Yaeta; Kumar, Penmetcha; Nishikawa, Shigemichi  
PATENT ASSIGNEE(S): National Institute of Advanced Industrial Science and Technology, Japan; Wakenyaku Co., Ltd.  
SOURCE: PCT Int. Appl., 42 pp.  
CODEN: PIXXD2  
DOCUMENT TYPE: Patent  
LANGUAGE: Japanese  
FAMILY ACC. NUM. COUNT: 1  
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
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WO 2002008443	A1	20020131	WO 2001-JP6226	20010718
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW				
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TG, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG				
CA 2416684	A1	20020131	CA 2001-2416684	20010718

AU 2001072748 A 20020205 AU 2001-72748 20010718  
 EP 1310564 A1 20030514 EP 2001-951912 20010718  
 R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE,  
 SI, LT, LV, FI, RO, MK, CY, AL, TR  
 AU 2001272748 B2 20060831 AU 2001-272748 20010718  
 US 20030162245 A1 20030828 US 2003-333417 20030317  
 US 7074595 B2 20060711

PRIORITY APPLN. INFO.:

JP 2000-220127 A 20000721  
 JP 2000-306119 A 20001005  
 WO 2001-JP6226 W 20010718

ED Entered STN: 01 Feb 2002

AB Selenomethionine-labeled protein is prepared with a wheat embryo cell-free wheat germ protein synthesis system by using selenomethionine-containing amino acids instead of methionine-containing amino acids, and protein synthesis under dialysis condition and batch conditions. Moreover, a process for producing a deuterium-labeled protein by using the same procedure was provided. Labeling of green fluorescent protein (GFP) and dihydrofolate reductase (DHFR) with the selenomethionine and deuterium was shown. The activity of the proteins was not altered by the labeling.

REFERENCE COUNT: 13 THERE ARE 13 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 98 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN

ACCESSION NUMBER: 2001:284088 CAPLUS [Full-text](#)

DOCUMENT NUMBER: 134:306148

TITLE: Cell-free protein synthesis using a modified mRNA having a TMV  $\Omega$  sequence or alfalfa mosaic virus leader sequence

INVENTOR(S): Endo, Yaeta

PATENT ASSIGNEE(S): Wakenyaku Co., Ltd., Japan

SOURCE: PCT Int. Appl., 82 pp.

CODEN: PIXXD2

DOCUMENT TYPE: Patent

LANGUAGE: Japanese

FAMILY ACC. NUM. COUNT: 1

PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2001027260	A1	20010419	WO 2000-JP7123	20001013
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CR, CU, CZ, DE, DK, DM, DZ, EE, ES, FI, GB, GD, GE, GH, GM, GR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MY, MZ, NZ, NI, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW				
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG				
CA 2385134	A1	20010419	CA 2000-2385134	20001013
AU 2000076861	A	20010423	AU 2000-76861	20001013
EP 1221481	A1	20020710	EP 2000-966474	20001013
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL				
JP 2005247857	A	20050915	JP 2005-78255	20050317
PRIORITY APPLN. INFO.:				
JP 1999-294370 A 19991015				
WO 2000-JP4814 A 20000718				
JP 2001-530465 A3 20001013				
WO 2000-JP7123 W 20001013				

ED Entered STN: 20 Apr 2001

AB Cell-free protein synthesis using a modified mRNA having a tobacco mosaic virus (TMV)  $\Omega$  sequence or alfalfa mosaic virus (AMV) leader sequence ligated



to the 5'-end and an untranslated region (UTR) ligated to the 3'-end, is disclosed. A plasmid for the RNA synthesis comprising a promoter and a terminator is claimed. A means for continuous cell-free protein synthesis using endospore-free wheat germ extract, where transcription and translation takes place sequentially, under dialysis, possibly using a porous filter, is claimed. Use of the cell-free protein synthesis method in evaluation of the relationship between genetic polymorphism and gene function, and screening of gene function, is claimed. Cell-free synthesis of various proteins, dihydrofolate reductase (dhfr), glutathione-S-transferase (GST), green fluorescent protein (GFP), GST-osteopontin fusion protein, luciferase, etc., is described.

REFERENCE COUNT: 10 THERE ARE 10 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 99 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN

ACCESSION NUMBER: 2000:814649 CAPLUS Full-text

DOCUMENT NUMBER: 133:331186

TITLE: Cell-free protein synthesis and preparation for cell-free protein synthesis

INVENTOR(S): Endo, Yaeta; Nishikawa, Shigemichi

PATENT ASSIGNEE(S): Wakenyaku Co., Ltd., Japan

SOURCE: PCT Int. Appl., 50 pp.

CODEN: PIXXD2

DOCUMENT TYPE: Patent

LANGUAGE: Japanese

FAMILY ACC. NUM. COUNT: 1

PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2000068412	A1	20001116	WO 1999-JP4088	19990729
W: AU, CA, CN, IL, KR, US, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE				
JP 2000316594	A	20001121	JP 1999-130393	19990511
JP 2000316595	A	20001121	JP 1999-130395	19990511
JP 2000333673	A	20001205	JP 1999-151599	19990531
CA 2373057	A1	20001116	CA 1999-2373057	19990729
AU 9949301	A	20001121	AU 1999-49301	19990729
AU 765632	B2	20030925		
EP 1176210	A1	20020130	EP 1999-933168	19990729
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, FI				
US 6905843	B1	20050614	US 2001-19995	20011120
US 20050186655	A1	20050825	US 2005-89652	20050325
US 7235382	B2	20070626		
JP 2005348739	A	20051222	JP 2005-203081	20050712
PRIORITY APPLN. INFO.:			JP 1999-130393	A 19990511
			JP 1999-130395	A 19990511
			JP 1999-151599	A 19990531
			WO 1999-JP4088	W 19990729
			US 2001-19995	A3 20011120

ED Entered STN: 21 Nov 2000

AB A preparation for synthesizing a cell-free protein which contains a cell extract prepared by eliminating inhibition system or inhibitors of self-protein synthesis; an apparatus for synthesizing a cell-free protein provided with a reaction tank for synthesizing the cell-free protein; and a kit to be used therefor are given. The above prepn. is obtained as a product which can be stored at room temperature while sustaining the biol. functions of the cell extract. A means for continuously synthesizing a cell-free protein comprising a cell extract from which substances inhibiting self-protein synthesis reactions have been substantially eliminated, involving a procedure selected

from among addition, preservation, exchange, and discharge of a factor selected from among mRNA template in the synthesis reactions, an enzyme associated with the energy regeneration system, a substrate, and an energy source. Preparation of luciferase and other protein with wheat germ extract pretreated with nonionic surfactant NP-40 and then subjected to tritin removal was shown.

REFERENCE COUNT: 10 THERE ARE 10 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 100 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN  
 ACCESSION NUMBER: 2000:616369 CAPLUS Full-text  
 DOCUMENT NUMBER: 133:173704  
 TITLE: Cell-free protein synthesis using wheat germ extract  
 INVENTOR(S): Endo, Yaeta  
 PATENT ASSIGNEE(S): Mitsubishi Chemical Corp., Japan; Cellfree Sciences Co., Ltd.  
 SOURCE: Jpn. Kokai Tokyo Koho, 10 pp.  
 CODEN: JKXXAF  
 DOCUMENT TYPE: Patent  
 LANGUAGE: Japanese  
 FAMILY ACC. NUM. COUNT: 1  
 PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
JP 2000236896	A	20000905	JP 1999-46379	19990224
JP 3753358	B2	20060308		
JP 2005312460	A	20051110	JP 2005-196673	20050705
PRIORITY APPLN. INFO.:			JP 1999-46379	A3 19990224

ED Entered STN: 06 Sep 2000

AB Embryo-free plant germ exts. are used for cell-free protein synthesis. The embryo-free plant germ exts. are free of endogenous inhibitors such as tritin and thionin. The plant germ is selected from wheat, barley, rice, and corn. The germ extract is prepared by sonication in the presence of surfactant and/or formycin-5'-phosphate. Amino acid, and energy source are supplied for the manufacture of proteins.

L7 ANSWER 101 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN  
 ACCESSION NUMBER: 2000:421640 CAPLUS Full-text  
 DOCUMENT NUMBER: 133:330997  
 TITLE: The construction of highly efficient cell-free protein synthesis from wheat embryos  
 AUTHOR(S): Endo, Yaeta  
 CORPORATE SOURCE: Japan  
 SOURCE: Bio Industry (2000), 17(5), 20-27  
 CODEN: BIINEG; ISSN: 0910-6545  
 PUBLISHER: Shi Emu Shi  
 DOCUMENT TYPE: Journal; General Review  
 LANGUAGE: Japanese

ED Entered STN: 23 Jun 2000

AB A review with 14 refs. on the cell-free protein synthesis with wheat germ. It comprises optimization of cell-free protein synthesis, instability and low translation initiation in the cell-free system, and development of high-efficiency cell-free system from wheat germ...

L7 ANSWER 102 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN  
 ACCESSION NUMBER: 1992:610746 CAPLUS Full-text  
 DOCUMENT NUMBER: 117:210746

ORIGINAL REFERENCE NO.: 117:36393a,36396a  
 TITLE: Cell-free, continuous synthesis of polypeptide  
 INVENTOR(S): Yokoyama, Shigeyuki; Endo, Yae; Kikawa, Takanori  
 PATENT ASSIGNEE(S): Japan  
 SOURCE: Jpn. Kokai Tokkyo Koho, 7 pp.  
 CODEN: JKXXAF  
 DOCUMENT TYPE: Patent  
 LANGUAGE: Japanese  
 FAMILY ACC. NUM. COUNT: 1  
 PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
JP 04200390	A	19920721	JP 1990-334103	19901130
PRIORITY APPLN. INFO.: ED Entered STN: 28 Nov 1992			JP 1990-334103	19901130

AB A cell-free, ribosome-containing, continuous system for synthesis of polypeptide (excluding chem. synthesis) from mRNA is disclosed. In this system, the reaction substrates, e.g., ATP, GTP, amino acids, etc., are continuously supplied to the reaction chamber having minimal air content and the reaction mixture is pumped through an ultrafiltration apparatus to sep. polypeptide products from the low-mol. substrates. The process minimizes foaming and therefore protein denaturation as compared to that of prior art. Synthesis of chloramphenicol acetyltransferase (CAT) was performed continuously for 17 h with a good yield.

L7 ANSWER 103 OF 103 CAPLUS COPYRIGHT 2008 ACS on STN

ACCESSION NUMBER: 1979:166178 CAPLUS Full-text

DOCUMENT NUMBER: 90:166178

ORIGINAL REFERENCE NO.: 90:26371a,26374a

TITLE:  $\alpha$ -Fetoprotein synthesis in yolk sac tumor: sex-dependent production of  $\alpha$ -fetoprotein by transplantable rat yolk sac tumor

AUTHOR(S): Endo, Y.; Kaneko, Y.; Urano, Y.; Tsuchida, Y.; Watabe, H.; Tsukada, Y.; Sakashita, S.; Hirai, H.; Oda, T.

CORPORATE SOURCE: Dep. Med., Univ. Tokyo, Tokyo, Japan

SOURCE: Scandinavian Journal of Immunology, Supplement (1978), 8(Carcinoembryonic Proteins: Recent Prog.), 181-6

CODEN: SJISDK; ISSN: 0301-6323

DOCUMENT TYPE: Journal

LANGUAGE: English

ED Entered STN: 12 May 1984

AB Expts. at cellular and subcellular levels were carried out. In cell incubation studies, yolk sac tumor cells maintained in female rat (YST-F cells) synthesized more  $\alpha$ -fetoprotein (AFP) than yolk sac tumor cells maintained in male rat (YST-M cells). AFP production was studied in cell-free protein-synthesizing systems derived from wheat germ, using tumor RNA. In this system, cytosol RNA from both YST-F and YST-M cells directed AFP synthesis. But the amount of AFP synthesized was smaller in the presence of RNA from YST-M cells. The reduced AFP synthesis by YST-M cells may be due to a quant. decrease in their cytosol mRNA coded for AFP.

=> s 16 AND ( freeze-dried OR freeze-drying OR freeze(2a)(dried OR drying) OR lyophiliz?)

L8 6 L6 AND (FREEZE-DRIED OR FREEZE-DRYING OR FREEZE(2A)(DRIED OR DRYING) OR LYOPHILIZ?)

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L8 ANSWER 1 OF 6 BIOSIS COPYRIGHT (c) 2008 The Thomson Corporation on STN  
ACCESSION NUMBER: 2006:589439 BIOSIS Full-text  
DOCUMENT NUMBER: PREV200600593047  
TITLE: Composition for cell-free protein synthesis.  
AUTHOR(S): Anonymous; Kuroita, Toshihiro [Inventor]; Kawakami, Bunsei  
[Inventor]; Kawamura, Yoshihisa [Inventor]; Nishikawa, Shigemichi [Inventor]; Endo,  
Yasuta [Inventor]  
CORPORATE SOURCE: Tsuruga, Japan  
ASSIGNEE: CellFree Sciences Co Ltd  
PATENT INFORMATION: US 07048915 20060523  
SOURCE: Official Gazette of the United States Patent and Trademark  
Office Patents, (MAY 23 2006)  
CODEN: OGPUPE7. ISSN: 0098-1133.  
DOCUMENT TYPE: Patent  
LANGUAGE: English  
ENTRY DATE: Entered STN: 8 Nov 2006  
Last Updated on STN: 8 Nov 2006  
ED Entered STN: 8 Nov 2006  
Last Updated on STN: 8 Nov 2006  
AB The present invention provides a composition for cell-free protein synthesis,  
which is superior in storage stability in a freeze-dried state, more  
particularly a freeze-dryable or freeze-dried composition for cell-free  
protein synthesis, which contains a cell extract for cell-free protein  
synthesis and inositol, and a freeze-dryable or freeze-dried composition for  
cell-free protein synthesis containing a cell extract for cell-free protein  
synthesis, and a deliquescent material in a proportion of not more than 0.01  
part by weight per part by weight of a protein in the composition; and a  
composition for cell-free protein synthesis superior in storage stability in a  
frozen state, more particularly a freezable or frozen composition for cell-  
free protein synthesis, containing a cell extract for cell-free protein  
synthesis and polyhydric alcohol.

L8 ANSWER 2 OF 6 CAPLUS COPYRIGHT 2008 ACS on STN  
ACCESSION NUMBER: 2007:428031 CAPLUS Full-text  
DOCUMENT NUMBER: 146:375315  
TITLE: Freeze-dried template for cell-free protein synthesis and  
laboratory application  
INVENTOR(S): Endo, Yasuta; Sawasaki, Tatsuya; Tanaka, Michihiro;  
Morishita, Akira; Saeki, Mihoro  
PATENT ASSIGNEE(S): Cellfree Sciences Co., Ltd., Japan  
SOURCE: Jpn. Kokai Tokkyo Koho, 27pp.  
CODEN: JKXXAF  
DOCUMENT TYPE: Patent  
LANGUAGE: Japanese  
FAMILY ACC. NUM. COUNT: 1  
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
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JP 2007097438	A	20070419	JP 2005-288613	20050930
PRIORITY APPLN. INFO.:			JP 2005-288613	20050930

ED Entered STN: 19 Apr 2007

AB The freeze-dried template is nucleotide sequence for visible proteins selected  
from green fluorescent protein, blue fluorescent protein, etc. The RNA  
polymerase and phosphotransferase such as creatine kinase used in the cell-  
free protein synthesis do not contain animal and microbial contaminants. With

proper solns., the freeze-dried template-containing cell-free protein synthesis system may be easily used, and protein synthesis monitored by the presence of visible proteins. Also, the cell-free protein synthesis system such as wheat germ extract does not use expansive RNase inhibitors.

L8 ANSWER 3 OF 6 CAPLUS COPYRIGHT 2008 ACS on STN  
 ACCESSION NUMBER: 2003:913316 CAPLUS Full-text  
 DOCUMENT NUMBER: 139:360695  
 TITLE: Lyophilized preparation for cell-free protein synthesis  
 INVENTOR(S): Endo, Yaeta; Ogasawara, Tomio  
 PATENT ASSIGNEE(S): Japan  
 SOURCE: PCT Int. Appl., 32 pp.  
 CODEN: PIXXD2  
 DOCUMENT TYPE: Patent  
 LANGUAGE: Japanese  
 FAMILY ACC. NUM. COUNT: 1  
 PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2003095661	A1	20031120	WO 2003-JP5656	20030506
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NI, NO, NZ, OM, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PT, RO, SE, SI, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG AU 2003235844 A1 20031111 AU 2003-235844 20030506 CA 2485827 A1 20031120 CA 2003-2485827 20030506 EP 1550728 A1 20050706 EP 2003-721030 20030506 R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR, BG, CZ, EE, HU, SK US 20050153390 A1 20050714 US 2004-514855 20041221 PRIORITY APPLN. INFO.: JP 2002-138828 A 20020514 WO 2003-JP5656 W 20030506				

ED Entered STN: 21 Nov 2003

AB The lyophilized cell-free protein synthesis system has comparable activity to that prepared by low-temperature preservation. The cell-free protein synthesis system contains lower deliquescent substances such as potassium acetate. The deliquescent substances amount to  $\leq 0.01$  weight% based on 1 weight% protein in the cell-free protein synthesis system. The low-mol. weight protein synthesis inhibitors are removed in the presence of high-energy phosphate compds. such as ATP.

REFERENCE COUNT: 9 THERE ARE 9 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L8 ANSWER 4 OF 6 CAPLUS COPYRIGHT 2008 ACS on STN  
 ACCESSION NUMBER: 2003:836518 CAPLUS Full-text  
 DOCUMENT NUMBER: 139:319681  
 TITLE: Composition for cell-free protein synthesis  
 INVENTOR(S): Kuroita, Toshihiro; Kawakami, Bunsei; Kawamura, Yoshihisa; Nishikawa, Shigemichi; Endo, Yaeta  
 PATENT ASSIGNEE(S): Toyo Boseki Kabushiki Kaisha, Japan; Wakenyaku Co., Ltd.; Cellfree Sciences Co., Ltd.  
 SOURCE: U.S. Pat. Appl. Publ., 20 pp.  
 CODEN: USXXCO

DOCUMENT TYPE: Patent  
LANGUAGE: English  
FAMILY ACC. NUM. COUNT: 1  
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
US 20030199076	A1	20031023	US 2002-124953	20020418
US 7048915	B2	20060523		

PRIORITY APPLN. INFO.: US 2002-124953 20020418

ED Entered STN: 24 Oct 2003

AB The present invention provides a composition for cell-free protein synthesis, which is superior in storage stability in a freeze-dried state, more particularly a freeze-dryable or freeze-dried composition for cell-free protein synthesis, which contains a cell extract for cell-free protein synthesis and inositol, and a freeze-dryable or freeze-dried composition for cell-free protein synthesis containing a cell extract for cell-free protein synthesis, and a deliquescent material in a proportion of not more than 0.01 part by weight per part by weight of a protein in the composition; and a composition for cell-free protein synthesis superior in storage stability in a frozen state, more particularly a freezable or frozen composition for cell-free protein synthesis, containing a cell extract for cell-free protein synthesis and polyhydric alc.

REFERENCE COUNT: 6 THERE ARE 6 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L8 ANSWER 5 OF 6 CAPLUS COPYRIGHT 2008 ACS on STN

ACCESSION NUMBER: 2002:341319 CAPLUS Full-text

DOCUMENT NUMBER: 136:336852

TITLE: Stabilization of freeze-dried cell extract for cell-free protein synthesis

INVENTOR(S): Kuroita, Toshihiro; Kawakami, Fumikiyo; Kawamura,

Yoshihisa; Nishikawa, Shigemichi; Endo, Yaeta

PATENT ASSIGNEE(S): Toyobo Co., Ltd., Japan; Wakenyaku Co., Ltd.

SOURCE: Jpn. Kokai Tokkyo Koho, 14 pp.

CODEN: JKXXAF

DOCUMENT TYPE: Patent

LANGUAGE: Japanese

FAMILY ACC. NUM. COUNT: 1

PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
JP 2002125693	A	20020508	JP 2000-320106	20001019
			JP 2000-320106	20001019

PRIORITY APPLN. INFO.:

ED Entered STN: 08 May 2002

AB Inositol 0.3-3 weight% and polyalcs. 0.1-10, based on the weight of the protein in the cell ext., are useful for stabilization of freeze-dried cell extract for cell-free protein synthesis. The cell extract is obtained from wheat, barley, spinach, reticulocyte, Escherichia coli, etc. The cell extract may contain physiol. active substance selected from creatine kinase, pyruvate kinase, RMA polymerase, and chaperone proteins.

L8 ANSWER 6 OF 6 CAPLUS COPYRIGHT 2008 ACS on STN

ACCESSION NUMBER: 2000:814649 CAPLUS Full-text

DOCUMENT NUMBER: 133:331186

TITLE: Cell-Free protein synthesis and preparation for cell-free protein synthesis

INVENTOR(S): Endo, Yaeta; Nishikawa, Shigemichi

PATENT ASSIGNEE(S): Wakenyaku Co., Ltd., Japan  
 SOURCE: PCT Int. Appl., 50 pp.  
 CODEN: PIXXD2  
 DOCUMENT TYPE: Patent  
 LANGUAGE: Japanese  
 FAMILY ACC. NUM. COUNT: 1  
 PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2000068412	A1	20001116	WO 1999-JP4088	19990729
W: AU, CA, CN, IL, KR, US, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE				
JP 2000316594	A	20001121	JP 1999-130393	19990511
JP 2000316595	A	20001121	JP 1999-130395	19990511
JP 2000333673	A	20001205	JP 1999-151599	19990531
CA 2373057	A1	20001116	CA 1999-2373057	19990729
AU 9949301	A	20001121	AU 1999-49301	19990729
AU 765632	B2	20030925		
EP 1176210	A1	20020130	EP 1999-933168	19990729
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, FI				
US 6905843	B1	20050614	US 2001-19995	20011120
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PRIORITY APPLN. INFO.:				
			JP 1999-130393	A 19990511
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			JP 1999-151599	A 19990531
			WO 1999-JP4088	W 19990729
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ED Entered STN: 21 Nov 2000

AB A preparation for synthesizing a cell-free protein which contains a cell extract prepared by eliminating inhibition system or inhibitors of self-protein synthesis; an apparatus for synthesizing a cell-free protein provided with a reaction tank for synthesizing the cell-free protein; and a kit to be used therefor are given. The above prep. is obtained as a product which can be stored at room temperature while sustaining the biol. functions of the cell extract. A means for continuously synthesizing a cell-free protein comprising a cell extract from which substances inhibiting self-protein synthesis reactions have been substantially eliminated, involving a procedure selected from among addition, preservation, exchange, and discharge of a factor selected from among mRNA template in the synthesis reactions, an enzyme assocd. with the energy regeneration system, a substrate, and an energy source. Preparation of luciferase and other protein with wheat germ extract pretreated with nonionic surfactant NP-40 and then subjected to tritin removal was shown.

REFERENCE COUNT: 10 THERE ARE 10 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

=> d his

(FILE 'HOME' ENTERED AT 14:59:34 ON 04 JUN 2008)

FILE 'MEDLINE, BIOSIS, CAPLUS, EMBASE' ENTERED AT 14:59:49 ON 04 JUN 2008

E ENDO Y/AU  
 L1 2996 S E3-E7  
 E SAWASAKI T/AU  
 L2 91 S E3  
 L3 202 S E11-E12

L4 3093 S L1-L3  
L5 215 S L4 AND ((PROTEIN(2A)SYNTHES?) OR TRANSLATION) (3A) ((IN(A)VITRO) OR  
CELL(A)FREE OR CELL-FREE)  
L6 121 DUP REM L5 (94 DUPLICATES REMOVED)  
L7 103 S L6 AND (MRNA OR TEMPLATE OR WHEAT OR ATP)  
L8 6 S L6 AND ( FREEZE-DRIED OR FREEZE-DRYING OR FREEZE(2A) (DRIED OR  
DRYING) OR LYOPHILIZ?)

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